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RELIABILITY OF LABORATORY-BRED STOCKS AS SAMPLES OF WILD POPULATIONS, AS SHOWN IN A STUDY OF THE VARIATION OF *PEROMYSCUS POLIONOTUS* IN PARTS OF FLORIDA AND ALABAMA*

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INTRODUCTION

POPULATIONS of *Peromyscus polionotus* have a high degree of variability throughout the range of the species and consequently the group offers unusual opportunities for the study of natural variation. Seven subspecies have been described from Florida and parts of adjacent states. The animals considered in the present study are from parts of northwestern Florida and southern Alabama, where breeding stocks were collected in the spring of 1935 (Fig. 1).

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The mice on the pale sandy beaches of the Gulf coast east of Pensacola, Florida, and upon the light sands, which have a gray or buff hue for a number of miles inland, have been described as *Peromyscus polionotus albifrons* Osgood. North and farther inland from this region, upon heavier and darker soils, are darker mice which have been named *Peromyscus polionotus polionotus* (Wagner). A third race, *Peromyscus polionotus leucocephalus* Howell, occurs only on Santa Rosa Island, a long and narrow strip of sand lying in the Gulf of Mexico, close to shore (Osgood, 1909; Howell, 1920, 1921; Sumner, 1926). Members of these three races seem to occupy similar ecological positions within their respective ranges. They burrow in open areas, often in fields no longer cultivated or in beach sands; hence the common names ''oldfield mouse'' and ''beach-mouse.''

That these mice are protectively colored in at least a general manner seems apparent. F. S. Sumner has discussed the habits, the distribution, and especially the variability among these mice. In his earlier papers (1929, 1929*a*) he was inclined to minimize the importance of soil color in relation to pelage color, but later (1934) he admitted a ''causal relation.'' No measurements of the actual soil color appear to have been made by him. Cott (1940) has discussed Sumner's findings in this area as an example of ''concealment in defense,'' and Huxley (1942) has described the situation as being the result of natural selection.

One objective of the present study was to examine the apparent relationship between pelage color and soil color, especially to determine if possible whether local populations were adjusted in pelage color to the precise hue of the local soil. Also considered of importance was the question of the variability within local populations and the sources of variation which contribute to the observed total variability.

ACKNOWLEDGMENTS

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SOURCE OF THE DATA

The specimens of mice measured for this study were prepared from laboratory-reared animals. Many of the characteristics measured change with age and therefore it was desirable to use animals of known age. Since there are no satisfactory criteria for determining the ages of wild-caught mice of the genus *Peromyscus,* the only method of obtaining mice of known age is to raise them in a laboratory. In addition, when differences between mice from two localities persist in the laboratory-bred offspring, the differences can be assumed to be inherited and not directly induced by some environmental factor.

The wild-caught parents of the laboratory stocks were all trapped by myself. The mice were captured at the several collecting stations and shipped to the Laboratory of Vertebrate Biology at Ann Arbor, Michigan. Here the animals were mated and the young reared according to a standardized technique, fully described by Dice (1929, 1932). All of the animals were kept under the same laboratory conditions. Because of reduced fertility and increased mortality, certain stocks took longer than others to produce series of one-year-old animals. The slower breeding stocks were, therefore, in captivity for some time longer than the others. As far as is known, the conditions during this additional period continued to be the same as earlier. Not all of the wild mice sent to the laboratory were represented in the final data, since some produced no offspring. The heredity of a few of the wild mice may be represented in more than one sibship in the final assembly of data, because some of their offspring may have been mated with other mice of the same stock to increase the number of breeding animals. This has been allowed for in making the computations. In some stocks, the samples of the wild population are much less full than would be desirable, due partly to the above-mentioned infertility and mortality.

The one-year age class of mice was limited to animals from thirty-six to seventy-eight weeks of age. The two-year age class contained mice older than seventy-eight weeks. No mice younger than thirty-six weeks have been considered in this study. All comparisons were based upon one-year-old animals, except in the Santa Rosa Island data, in which two-year-old animals, were also used, but only for color-measurement data.

COLLECTING LOCALITIES

The wild-caught parents of the laboratory stocks were secured at the following collecting localities (Fig. 1):

SANTA ROSA ISLAND.—The animals were captured on the beach and along a line of low dunes on the south (Gulf) side of Santa Rosa Island, about two and a half miles east of the bridge at Fort Walton, in Escambia County, Florida. Almost the entire soil surface was exposed, the only vegetation consisting of widely scattered coarse grass. The soil was a very light-colored, almost white, sand. The island has been described by Sumner (1926, 1929). Sixteen animals were sent to the laboratory. Six wild-caught parents are represented by five sibships with thirty-eight one-year-old and two-year-old offspring.

Destin.—On the beach about two miles east of the small town of Destin, Okaloosa County, Florida. Destin lies at the tip of a narrow peninsula of the mainland, just east of Santa Rosa Island. This location is probably a few miles west of that termed East Pass by Sumner. The exposed beach at this point was made up of drifting white sand, anchored in small mounds or dunes by shrub roots. The beach itself was about one hundred yards wide. It gave way suddenly to a very dense growth of shrubs. According to field notes, footprints and burrows of the mice extended well back into these shrubs. The soil on the beach was almost entirely exposed. Twenty-six animals were sent in, resulting in thirty-three one-year-old mice in seven sibships, descended from nine wild-caught parents.

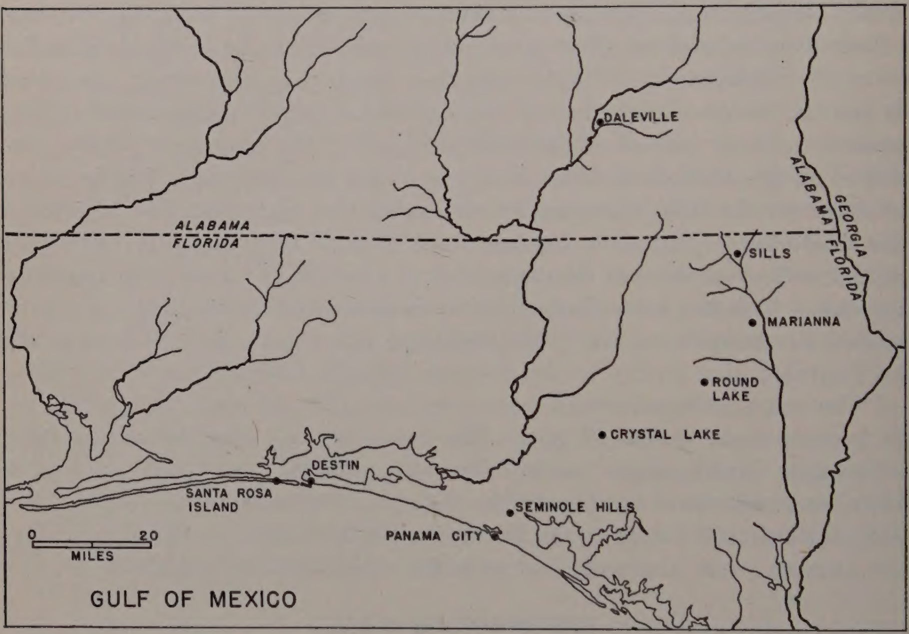

Fig. 1. A portion of northwestern Florida and southern Alabama, showing the collecting localities of this study.

Panama City.—Along a line of dunes, immediately above the beach, about nineteen miles west of Panama City, Bay County, Florida. The vegetation on the dunes was scattered, with several large closely growing clumps of shrubs. Back of the dunes was a dense growth of shrubs of uniform height. A search of this scrub area did not reveal any signs of *polionotus* activity. The mice seemed to be confined to the more open dunes and beach.

The beach sands were white, very similar in hue to those at Destin and Santa Rosa Island. This white sand extended about a half mile inland and gave way suddenly at that point to the gray or buff soil characteristic of the flatwoods. At the same distance from the beach was a narrow zone of poorly drained, swampy land. No field notes were made on the extent of this belt of wet land behind the beaches, except to record its presence at the Panama City location. Apparently, Kurz (1942) found a similar development of a ''flatwoods swale'' back of the beach in the same county, at Beacon Hill, Florida, and both Martens (1931) and Vernon (1942) mentioned the effect of the beach ridge in determining the drainage pattern behind the beaches along this particular stretch of Florida coast line.

Fourteen animals were sent to the laboratory from Panama City; nine of these are represented by six sibships containing twenty-three one-year-old mice.

SEMINOLE HILLS.—About five miles northwest of the town of Westbay, Walton County, Florida, and about six or seven miles directly north of the last location (Panama City). The country here was pine flatwoods, largely cut over and burned over at various times. The ground cover in the drier areas was of scattered clumps of wire grass which allowed a good part of the soil to show. Where there was more moisture, the growth of vegetation was denser and completely covered the soil. The mice were taken at the edge of a formerly cultivated field lying in the flatwoods. A soil sample was taken in the flatwoods near by, for the soil of the field itself was somewhat more yellow owing to a different-colored soil brought up by tillage. Twenty-two mice were sent in, resulting in eighty-seven one-year-old mice in fourteen sibships, descended from sixteen wild-captured animals.

CRYSTAL LAKE.—Near the town of Crystal Lake, Washington County, Florida, in the area described by Harper (1914) as the West Florida lake region. Most of the vegetation was of an open pine-wood type. Sinks were very common. Some oaks were present throughout the pine woods, and in the lower, richer spots hammocks of live oak and other trees had been formed. The grass cover was rather thin, exposing a good part of the soil. Fires had been common. Thirteen mice were sent in, twelve of which are represented in twenty-one sibships containing ninety-three one-year-old mice.

ROUND LAKE.—An abandoned field about two miles south of the town of Round Lake, Jackson County, Florida. The uncultivated land was covered with a growth of small oaks in a fairly open stand with grass beneath the trees. The grass cover was uniform but rather open, so that the soil color was exposed. According to field notes, Round Lake lay about at the northern edge of a region of lighter-colored soil. Northward of Round Lake the soil varied in color; some areas were red and others paler, but the red soil became more and more frequent toward the north. South of Round Lake the soil was rarely red or dark. Fifteen mice were sent in, and twelve of these are represented in eight sibships containing seventy-eight one-year-old mice.

MARIANNA.—In an old field at the west edge of the city of Marianna, Jackson County, Florida. The ground was covered by a uniform growth of grass and weeds, although the soil color could be seen at almost any spot because of the thinness of the vegetation. Much of the land not in cultivation within recent years was wooded. In some places the soil supported a thick stand of trees; elsewhere the stand was thin. The soil was dark reddish, in distinct contrast to that even a few miles south. Harper (1914) has described this area as the "Marianna Red Lands." Seven mice were sent in, and all seven are represented in the fifteen sibships containing 105 animals.

SILLS.—Eight miles east of the town of Campbellton, Jackson County, Florida, probably in the "Marianna Red Lands" of Harper (1914). The land had apparently once been mostly under forest, but had been cleared for the large part and was in cultivation. What forests remained, chiefly along swamps and beside rivers, were relatively heavy stands. Old fields were thick with grass and weeds, the color of the soil being exposed only occasionally. On some hillsides or eroded land the soil color showed somewhat more plainly. The mice were taken in a cultivated field of forty-five acres, completely surrounded by a heavily wooded swamp, the nearest open land being over a half mile away. Twenty mice were sent in, twelve of which are represented in nine sibships containing fifty-three one-year-old animals.

DALEVILLE.—Four miles southwest of Daleville, Dale County, Alabama. The region, like that at Sills, was predominantly one of cultivated land. Forests where present were heavy. The soil color was variable, some soils being gray, some light reddish brown like the sample taken, and other soils much darker. According to the classification of Harper (1928), this is in the "Eastern Red Hills" of Alabama. Field notes indicate that this general region was of a darker, redder soil than the sample taken. Twenty-five mice were sent in, eight of which produced eight sibships with thirty-two one-year-old animals.

SOIL COLORS

The soil sample from Daleville is reddish brown, as indicated by the sample of the surface soil read by the tint photometer (Table I). There are pronounced differences among the soils in both shade and hue.

Santa Rosa Island is entirely covered with white sand. The sands of the mainland beaches, east of Santa Rosa Island, are almost as pale. The readings of the sample from Destin are almost the same as those of the sample from Santa Rosa Island (Table I). The sand from Panama City is only slightly darker, to judge from the tint-photometer readings.

The sample from Seminole Hills, darker than samples from the beaches, is gray, and the readings through the green and blue-violet screens are only

TABLE I

COLOR OF SURFACE SOILS

Average of ten tint-photometer readings of different parts of one soil sample from each collecting locality.

Collecting Location	Red	Green	Blue-violet
Santa Rosa Island	65.0	61.0	58.0
Destin	66.5	61.6	57.2
Panama City	61.0	59.6	59.6
Seminole Hills	28.5	26.2	24.0
Crystal Lake	26.9	19.6	14.6
Round Lake	31.0	23.2	18.4
Marianna	22.0	10.2	7.0
Sills	22.4	12.2	7.5
Daleville	28.5	26.2	15.4

slightly below the reading through the red screen. The Crystal Lake sample has a reading for red which is almost as high as that of the Seminole Hills soil, but the readings through the green and the blue-violet screens are much lower, indicating that the Crystal Lake sample is darker and more reddish. The Round Lake soil is buff, with a relatively high reading for red, and lesser though not greatly reduced readings for green and blue-violet.

The soil samples from Marianna and Sills are very similar both in appearance and in tint-photometer readings. The relatively high readings for red and the low readings for green and blue-violet indicate that these samples are dark red.

The soil sample from Daleville is reddish brown, as indicated by the relatively high readings for red and for green, and the lower reading for blue-violet.

Between the white beach sands and the inland gray flatwoods soils the shift in soil color is sudden. As observed at Panama City, the change came about a half mile behind the beach. In contrast, the change from the lighter buff or gray soils to the dark red soils, as observed between Round Lake and Marianna, was not sudden and complete, but consisted rather of areas of both soil colors interspersed over a distance of a number of miles.

METHODS OF MEASUREMENT

All external linear measurements of the mice were made by me immediately after the mice were killed. The method of making these measurements has been described by Dice (1932) and does not seem to differ significantly from that described by Sumner (1927). Sumner's animals were field-caught, however, and in captivity nine to eleven months (Sumner, 1929); in the present study only one-year-old laboratory-bred animals were used in comparing linear measurements.

Readings of the pelage color have been made by a standardized technique (Dice, 1932). As differences in color readings in stocks taken in approximately the same locality by Sumner and myself are explained, at least in part, by differences in technique, these differences will be reviewed.

In both studies the color of the pelage was measured by means of the Ives tint photometer. This instrument was used by Sumner in his *polionotus* study, and its construction and method of operation were described by him (1927). Certain modifications were adopted by Dice (1932). The tint photometer is the same type of instrument as used by Sumner, but a pair of lenses has been inserted to restrict the field of view to a smaller area. Both the skin and the magnesium carbonate block are covered by a black plate of heavy brass, with two rectangular apertures. These apertures are somewhat larger than the fields of view allowed by the lenses of the machine. Beside holding the skins flat, this black brass plate reduces to a minimum the extraneous light reflected from the parts of the skin and parts of the magnesium carbonate block not included in the field of vision of the machine. Sumner used a glass plate over the pelage and over the block, and its use is said by him to have reduced color reading about 5 per cent. No glass plate was used in this study. In Sumner's studies the skin was illuminated from the side. The illumination in the present study is from the rear of the skin, with the nose of the flat skin being placed toward the machine.

The most important difference between these color-reading techniques is in the size and location of the small areas of pelage chosen for reading. In Sumner's studies the area read extended across the animal's back and included both the darker middorsal stripe and parts of the lighter sides. In my study, the field of view of the instrument was much smaller, and the dorsal reading could be restricted to the dorsal stripe. I made a second reading which was restricted chiefly to the side of the animal but included none of the white ventral pelage. Sumner's readings correspond, in a sense, to an average of the dorsal and the side readings of my study. The two sets of readings cannot, therefore, be compared directly.

STATISTICAL ANALYSIS OF THE MEASUREMENTS

Summaries of the measurements for body and skeleton for the several stocks are given in Table II and for pelage color in Table III. These tables list for each characteristic studied, in each stock of mice, the values of the stock mean and its standard error, the extremes of individual measurements, the actual number and the effective number of sibships, and the number of individual laboratory-bred mice in stock. The method of computing these various statistics will be discussed in succeeding parts of this paper.

When stating the statistical significance of differences between means in this paper, the conventional terms "significant" and "highly significant" will be used to designate differences significant at, respectively, the 5 per

TABLE II

Body and Skeleton Measurements in *Peromyscus polionotus* from Florida and Alabama
Means, standard errors, and extremes in millimeters. Means and standard errors are computed from sibship means.

Stock	Number of Laboratory Offspring	Number of Sibships		Body Length	Tail Length	Hind-foot Length	Ear Length	Femur Length	Mandible Length	Condyle-premaxilla Length	Condyle-zygoma Length	Bullar Width
		Actual	Effective									
Santa Rosa Island	28	5	3.0	77.61 ± 1.24 72–83	53.60 ± 1.90 43–61	18.65 ± .38 17.8–20.0	15.12 ± .25 13.8–16.0	14.77 ± .20 13.7–15.9	14.44 ± .07 13.9–15.4	22.26 ± .14 21.2–23.3	16.54 ± .08 15.8–17.4	10.36 ± .06 9.9–10.9
Destin	24	3	3.0	77.99 ± 1.42 75–84	56.99 ± 1.05 50–62	18.15 ± .19 17.0–18.8	16.14 ± .11 15.0–17.1	14.76 ± .06 13.8–15.3	14.53 ± .19 13.8–15.4	22.14 ± .16 21.0–22.7	16.65 ± .07 16.0–17.0	10.15 ± .06 9.7–10.5
Panama City	9	2	1.5	75.54 ± 1.40 72–78	51.62 ± 3.61 48–56	17.56 ± .13 17.1–18.5	15.56 ± .68 14.6–17.0	14.18 13.9–14.4	14.32 14.1–14.4	21.95 21.6–22.1	16.45 16.2–16.6	9.95 9.9–10.1
Seminole Hills	63	7	5.0	73.26 ± 1.88 63–81	48.31 ± .93 39–56	16.62 ± .18 15.8–17.9	14.66 ± .16 13.6–15.8	13.56 ± .16 12.4–15.2	13.46 ± .09 12.9–14.4	20.89 ± .16 20.0–22.2	15.6 ± .10 14.9–16.5	9.73 ± .07 8.9–10.2
Crystal Lake	58	9	4.5	76.02 ± .83 69–81	50.05 ± .80 45–58	16.67 ± .16 15.5–18.0	15.53 ± .24 13.9–17.3	13.61 ± .25 12.6–15.1	13.69 ± .17 12.8–14.8	21.14 ± .21 20.0–22.2	15.71 ± .17 14.8–16.6	9.92 ± .12 9.4–10.4
Round Lake	78	8	5.5	73.52 ± .47 69–80	47.11 ± 1.24 40–54	16.66 ± .18 15.5–17.6	15.02 ± .34 14.0–16.1	13.41 ± .12 12.7–14.2	13.82 ± .06 13.1–14.4	21.23 ± .14 20.2–22.0	15.69 ± .08 14.9–16.3	9.93 ± .05 9.5–10.3
Marianna	90	10	3.5	76.99 ± 1.14 69–84	50.36 ± .79 45–56	17.00 ± .18 16.0–18.0	15.99 ± .27 14.8–17.1	14.10 ± .17 13.0–15.4	14.28 ± .09 13.5–14.9	21.68 ± .12 20.8–22.6	16.08 ± .10 15.2–16.6	10.17 ± .08 9.7–10.5
Sills	46	6	5.0	74.53 ± 1.40 69–80	49.18 ± 1.30 43–55	16.81 ± .22 15.7–17.8	15.66 ± .18 14.5–16.4	13.78 ± .17 12.4–14.8	13.93 ± .16 13.0–14.6	21.37 ± .23 20.0–22.4	15.81 ± .16 15.1–16.5	10.08 ± .13 9.5–10.3
Daleville	23	4	3.0	78.94 ± .99 70–83	50.98 ± 1.44 44–56	16.78 ± .22 16.0–18.0	15.92 ± .21 14.0–17.0	14.35 ± .14 13.0–15.2	14.34 ± .05 13.7–14.7	21.82 ± .12 20.4–22.5	16.38 ± .16 15.1–17.0	10.41 ± .11 9.8–10.7

cent level and the 1 per cent level. Any difference beyond the 5 per cent level will be termed "not significant."

The statistical calculations were directed toward obtaining two estimates, the first of the means of the measurements for that stock, and the second of the variability of the measurements. It is assumed in this, as in similar studies of variability, that these estimates, derived from the data at hand, are related in some definite manner to the wild population from which the parents of the animals were drawn. Just how close this correspondence may be between the sample population as raised in the laboratory and the wild population depends upon a number of factors, including the size of the sample from the wild population, the mode of inheritance of the qualities studied, and the number of individuals at hand on which to base the estimate.

In a number of the studies made in the past, it has been the practice in the Laboratory of Vertebrate Biology to rear a number of offspring from wild-caught parents, avoiding inbreeding as much as possible, and to base the computed values of mean, standard deviation, and standard error upon the distribution of the measurements of the one-year-old offspring, treated as one population. Dice (1932) early called attention to the dangers of an inadequate sampling of the wild population by this method.

More recently, there has been an attempt to define the significance of differences between means in biological as well as in statistical terms. Hubbs and Perlmutter (1942) have suggested that when two population means differ by an amount equal to the sum of their respective standard deviations, such a difference is "approximately of the order that may be utilized for subspecific separation." Dice (1944) has further discussed the usefulness of the standard deviation in judging the extent of divergence between populations.

These authors seem to agree that in their experience, when comparing the means of almost any two populations a statistically significant difference may be demonstrated, provided that large enough samples are obtained. Such differences may be very small, in fact may appear negligible in our understanding of ecological importance. Yet the fact that such differences are so often observed to be consistent upon further sampling seems to indicate that the differences, no matter how small, reflect biological differences of the same order. Where such biological differences are absent, as from two samples drawn from the same population, unlimited further sampling should suggest statistically significant differences between means only in the small proportion of cases defined by the confidence level adopted for statistical significance.

The criterion suggested by Hubbs and Perlmutter for defining biological significance is an arbitrary standard judged from their experience to be useful in systematic studies. Other fields may reasonably determine their own

TABLE III

Pelage Color Measurements in *Peromyscus polionotus* from Florida and Alabama

Means, standard errors, and extremes, in per cent. Means and standard errors are computed from sibship means.

Stock	Laboratory Offspring	Sibships		Effective Average Offspring per Sibship (k_o)	Dorsal Readings			Side-stripe Readings		
		Actual	Effective		Red	Green	Blue-violet	Red	Green	Blue-violet
Santa Rosa Island	21	4	2.5	4.2	18.93 ± .74 14–26	15.30 ± .82 12–21	13.90 ± .60 11–17	27.41 ± 1.89 20–35	22.57 ± 1.37 16–31	19.26 ± 1.79 13–28
Destin	33	7	4.5	3.3	14.11 ± .40 12–16	10.50 ± .20 9–12	8.93 ± .33 6–10	20.05 ± .93 15–22	14.91 ± .45 12–17	12.05 ± .46 10–15
Panama City	20	6	3.0	3.2	12.41 ± .48 11–15	9.53 ± .31 8–12	7.61 ± .29 6–10	18.17 ± 1.06 15–23	14.32 ± .60 11–18	11.53 ± .61 9–16
Seminole Hills	87	14	8.0	5.8	11.06 ± .28 9–14	8.58 ± .15 7–11	7.04 ± .26 5–9	16.91 ± .46 10–21	12.84 ± .34 7–16	10.12 ± .29 5–14
Crystal Lake	93	21	6.0	4.2	13.74 ± .33 11–20	9.82 ± .36 7–15	7.21 ± .36 5–12	20.30 ± 1.00 13–30	14.74 ± .78 8–21	11.60 ± .79 6–16
Round Lake	76	8	5.5	9.0	10.65 ± .22 8–15	8.12 ± .31 6–11	6.66 ± .39 4–10	16.97 ± .60 12–22	12.60 ± .54 7–15	10.14 ± .54 8–14
Marianna	103	15	3.5	6.7	8.76 ± .51 7–15	6.88 ± .31 5–11	5.50 ± .24 4–9	16.17 ± .95 12–24	12.64 ± .71 8–19	10.24 ± .69 7–15
Sills	53	9	6.0	5.3	7.71 ± .26 6–14	6.04 ± .19 5–10	5.23 ± .16 4–9	14.12 ± .38 11–16	10.83 ± .24 8–13	9.14 ± .44 7–11
Daleville	32	8	4.0	3.8	7.35 ± .86 5–11	5.48 ± .63 4–8	5.22 ± .62 4–8	12.34 ± .24 7–15	9.09 ± .32 6–11	7.49 ± .31 6–11

practical standards for judging biological significance. For example, consistent differences which are clearly too small for taxonomic recognition may still be of fundamental interest in a study of those inherited differences which may exist between near-neighboring populations.

W. Hovanitz (1942) has criticized a number of the past studies, specifically those of Dice (1939, 1940, 1941) and of Blair (1941), on the grounds that conclusions which are based upon a study of the variation in the offspring of a few wild-caught animals are misleading when applied to the wild population. He pointed out that the standard errors given in the studies quoted relate only to laboratory-bred stocks, not to wild populations.

In the present study an effort is made to determine the sources of variation and the true relation of the laboratory-bred stock to the wild population of which it is a sample.

The Mean

A true sample of the wild population of *Peromyscus polionotus* consists of wild-caught mice. Many of the characteristics studied, however, vary with the age of the individual and therefore cannot be measured in wild mice because of our ignorance of their age. Therefore, the wild-caught mice must be mated in pairs and offspring raised in order to measure the characteristics on mice of known age. This breeding procedure is essentially a second sampling (or subsampling), each sibship being a sample of the very large number of possible genetic combinations which might result from the mating of one pair of mice. The mathematical mean of the genotypes of the offspring will be the best available estimate of the mean of the genotypes of the two parents, assuming that some numerical value may be assigned to each genotype. The individual data are measurements of the phenotypic expression of the genotypes of the individual offspring. Dominance and epistasis as well as environmental interactions will determine this phenotypic expression.

There is no way of obtaining a direct measurement of the genetic composition of each item of the sample, that is to say, of each wild-caught mouse. Only by obtaining offspring from the mice may an estimate of the genetic make-up be obtained, and then only as an estimate of the combined effect of the two mice making up each pair. Thus, the average of a pair of wild-caught mice, as revealed by the average of their offspring, becomes the actual sampling unit. Consequently, the mean of the sibship means is the best available estimate of the mean of the wild population for the characteristic concerned.

Under ideal conditions, if a number of sibships were available, each containing the same number of mice, the unweighted mean of the sibship means would be identical with the general mean of all the laboratory-bred mice belonging to one stock. The general mean of the stock is, in reality, a weighted mean of sibship means, the weights being equal to the number of mice in each sibship. Where numbers are not uniform throughout the various sibships, however, this weighting seems improper, because there is no evidence that a more numerous laboratory sibship would necessarily resemble a larger part of the original wild population.

Thus, the mean used in past studies may depart from the best estimate of the wild mean, the amount of departure being governed by the number of sibships represented, and by the amount of uniformity throughout the sibships with regard to numbers of individuals in each. The range of possible error would seem to be minor where any considerable number of parent wild mice has been collected, decreasing as greater numbers of sibships and less variability in number of offspring per sibship prevent any unusual sibship from unduly affecting the mean.

When differences are present between the sexes as well as between sibships, the proper mean for the stock must take these differences into account. In this study the method of adjusting these differences is to compute each sibship mean as the average of means for the two sexes within that sibship. The stock mean is the average of all such means of sibships bred from parents taken at a particular geographic location.

The stock means used in the present study are all computed as unweighted means of the sibship means. In the body and skeletal measurements, where differences are found between the sexes, an average of the two sex means is used for each sibship mean. Consequently, in computing the mean of these measurements any sibship having only one sex represented cannot be used. In the color measurements, the mean of all animals within a sibship is used for the sibship mean, since color differences between the sexes are not apparent in these mice, or in any other mice of the genus *Peromyscus* which have been previously studied (Dice, 1932, 1933, 1937; Sumner, 1932; Osgood, 1909).

If the genetic make-up of all the parent mice were homozygous and identical, then there would be no inherited differences between sibships, and any one sibship would fully represent the single genetic type. Under such conditions it would be unnecessary to consider sibships as separate units, and there would be no advantage in using the average sibship means. But where the genetic constitution of the wild parents varies, as it evidently does in most of the stocks here studied, allowance must be made for that fact.

Measurement of Variability

Variability within a population is usually estimated by calculating the standard deviation or some other related measure of dispersion of the items about the mean. Such an index of variability may be used in two ways. First, the variability among individual items may be of interest in itself, to ascertain whether one stock may be more variable than another. Second, knowing the variability among individuals of a sample allows the estimation of the probable variability of sample means, which in turn may be the basis for judging the significance of differences between means of samples drawn from different wild populations. In the present study the variability must be estimated from the sibships, not from the original sample of wild-captured parents, just as the mean must be estimated from the same data.

Sibship and Sex Differences

Sibship differences in color readings were tested by a simple analysis of variance. Table IV gives the data pertaining to the Seminole Hills stock, as an illustration of the method. This stock consisted of fourteen sibships which together contained eighty-seven mice. The sibship means for the color reading used, the blue-violet, range from 6.8 to 8.7, while the individual measurements range from five to nine.

TABLE IV

ANALYSIS OF VARIANCE FOR DORSAL BLUE-VIOLET PELAGE COLOR READINGS IN *Peromyscus polionotus albifrons* FROM SEMINOLE HILLS

Source of Variability	Degrees of Freedom	Sums of Squares	Mean Square
Total	86	95.59	1.11
Between Sibships	13	46.52	3.58
Within Sibships	73	49.07	0.67

$$F = \frac{3.58}{0.67} = 5.33^{**}$$

** Highly significant.

The test for significance of differences between sibship means is made by dividing the mean square for "between sibships" by that "within sibships," and comparing the *"F"* ratio thus obtained with that to be found in a table of such values (Snedecor, 1946) for the numbers of degrees of freedom involved. In the present example, an F value of 5.33 shows that the sibship differences are highly significant, that is, that the chance of obtaining differences as large or larger due to sampling variation alone is less than one in a hundred.

Results of tests for sibship differences in all color readings in all stocks are shown in Table V. It is clear that for certain color readings in certain stocks there are highly significant differences between sibships, whereas for other colors, even within the same stocks, the differences may not be significant. In the Marianna stock, for example, highly significant differences exist in all side readings and in the dorsal red, while readings for dorsal green and blue-violet show no significant differences between sibships. The stocks from Santa Rosa Island and Destin, in contrast, show no significant differences between sibships in any of the color measurements.

TABLE V

ANALYSIS OF VARIANCE FOR COLOR MEASUREMENTS IN *Peromyscus polionotus*

Stock	Source of Variability	Degrees of Freedom	Mean Squares					
			Dorsal			Side		
			Red	Green	Blue-violet	Red	Green	Blue-violet
Santa Rosa Island	Total	20	4.09	5.69	3.03	24.63	22.56	18.36
	Between sibships	3	7.20	8.56	5.43	31.84	16.46	22.25
	Within sibships	17	3.54	5.18	2.61	23.36	23.64	17.68
Destin	Total	32	1.20	0.39	1.03	3.40	1.90	1.60
	Between sibships	6	2.68*	0.68	1.92*	4.56	1.28	1.31
	Within sibships	26	0.82	0.31	0.79	2.01	1.97	1.60
Panama City	Total	19	1.78	1.57	1.25	6.57	3.86	2.83
	Between sibships	5	1.52	1.04	0.69	6.59	3.04	3.79
	Within sibships	14	1.87	1.75	1.45	6.57	3.32	2.48
Seminole Hills	Total	86	1.17	0.61	1.11	4.57	2.71	2.05
	Between sibships	13	1.68	1.04*	3.58**	6.89	4.31	4.17**
	Within sibships	73	1.08	0.53	0.67	4.16	2.43	1.68
Crystal Lake	Total	92	2.62	2.09	2.52	14.18	6.75	7.82
	Between sibships	20	2.39	1.62	1.36	21.45*	11.28*	10.17
	Within sibships	72	2.68	2.22	2.71	12.17	5.49	7.16
Round Lake	Total	75	1.51	1.24	1.16	5.29	2.96	2.34
	Between sibships	7	2.81	4.22**	5.16**	12.55*	11.06**	12.02**
	Within sibships	68	1.38	0.93	0.74	4.54	2.13	1.34
Marianna	Total	102	1.96	1.16	0.86	5.18	3.49	3.44
	Between sibships	14	4.07**	1.54	1.14	13.71**	10.91**	10.26**
	Within sibships	88	1.63	1.10	0.82	3.82	2.31	2.36
Sills	Total	52	1.60	0.81	0.67	2.25	1.05	0.76
	Between sibships	8	1.79	1.13	0.58	5.17**	1.43	0.44
	Within sibships	44	1.56	0.75	0.68	1.72	0.98	0.82
Daleville	Total	31	2.08	1.03	1.35	2.64	1.19	1.16
	Between sibships	7	3.97	2.08*	2.47	1.06	2.08	1.60
	Within sibships	24	1.65	0.72	1.02	3.10	0.93	1.03

* Significant. ** Highly significant.

In the situation discussed above (Table IV and Table V), the data are classified in one way only, and a similar application of the analysis of variance can be made where the data are classified by sex alone or by sibship alone. In the color measurements, sex appears to be of no importance as a factor, so it can be disregarded and the data classified on the basis of sibship alone. Table VI shows the apparently random nature of the differences between sexes in the various sibships of the Round Lake stock.

TABLE VI

MEAN COLOR MEASUREMENTS FOR MALES AND FEMALES OF *Peromyscus polionotus albifrons* FROM ROUND LAKE

Sex and sibship means are based upon all animals in the particular subgroup.

	Sibships								Mean by Sexes
	1	2	3	4	5	6	7	8	
Males	7.5	10.0	8.4	8.4	7.0	7.3	7.3	9.0	8.1
Females	8.0	8.0	8.8	8.0	7.3	8.4	6.9	9.0	7.9
Sibship	7.8	9.0	8.6	8.2	7.3	8.0	7.1	9.0	—

In the body and bone measurements, the factor of sex proved to be important. Inspection of the data confirmed Sumner's (1926) observation that the females tend to be larger than the males in most measurements, although the two sexes do not seem to differ in foot length. Where the data must be classified both on the basis of sibship and on the basis of sex, the analysis of variance cannot here be applied directly since the numbers in the sex-sibship classes are not proportional. For example, sibship I (see Table VII) has two females to four males, but sibship IV is equally divided with ten of each sex. In sibship V the ratio is one female to eight males. For the analysis of variance to apply directly, the sex ratio must be identical in all sibships. This problem of disproportionate subclass numbers has been treated by Snedecor and Cox (1935) and Snedecor (1946), and their solution of the problem, entitled "Method of Expected Subclass Numbers," has been followed. Their method is applied in this study to only a part of the data relating to body and skeletal measurements. That part of the data chosen for examination includes all nine body and skeletal measurements for a single stock, Round Lake, but only two measurements, those for body length and hind-foot length, for all other stocks.

In the method of analysis used, a "primary" analysis of variance is carried out, using the various subclasses without regard to sex or sibship, each sex-sibship class being one subclass. The variability of individuals within these subclasses, being independent of the effect of differences between sexes or sibships, is assumed to be the best estimate of variability of individuals within classes, and is used to test the differences between sexes, or between sibships. In order to derive estimates either of means of

TABLE VII

ANALYSIS OF VARIANCE FOR BODY LENGTH, IN MILLIMETERS, IN *Peromyscus polionotus albifrons* FROM ROUND LAKE, BY METHOD OF EXPECTED SUBCLASS NUMBERS

Sibship	Data				Total
	Female		Male		
	Actual	Expected	Actual	Expected	
I. Number	2	2.46	4	3.54	6
Mean	75.00		69.50		
Sum		184.50		246.03	430.53
II. Number	1	.82	1	1.18	2
Mean	72.00		73.00		
Sum		59.04		86.14	145.18
III. Number	5	5.33	8	7.67	13
Mean	73.80		73.00		
Sum		393.35		559.91	953.26
IV. Number	10	8.20	10	11.79	20
Mean	76.30		73.70		
Sum		626.42		868.92	1495.34
V. Number	1	3.69	8	5.31	9
Mean	75.00		74.38		
Sum		276.75		394.96	671.71
VI. Number	3	3.28	5	4.72	8
Mean	73.40		71.00		
Sum		240.75		335.12	575.87
VII. Number	7	6.15	8	8.84	15
Mean	74.71		72.00		
Sum		459.47		637.20	1096.67
VIII. Number	3	2.05	2	2.95	5
Mean	76.70		73.00		
Sum		157.24		215.35	372.59
Total number	32	32.00	46	46.00	78
Mean					
Sum		2397.52		3343.63	5741.15

Primary Analysis of Variance

Source of Variability	Degrees of Freedom	Sums of Squares	Mean Square
Total	77	483.34	
Between Subclasses	15	187.93	
Within Subclasses	62	295.41	4.76

Analysis of Variance between Subclasses Using Expected Sums and Numbers

Source of Variability	Degrees of Freedom	Sums of Squares	
Total	15	223.08	
Between Sexes	1	97.27	
Between Sibships	7	107.94	
Sex × Sibship	7	17.86	

Completed Analysis of Variance

Source of Variability	Degrees of Freedom	Sums of Squares	Mean Square
Between Sexes	1	97.27	97.27**
Between Sibships	7	107.94	15.42**
Interaction (Sex × Sibship)	7	17.86	2.55
Error (within subclasses from primary analysis)	62		4.76

** Highly significant.

the various sibships or of means of the two sexes, corrected for disproportional numbers, a series of expected subclass numbers and sums is set up, and the analysis is continued, based upon these expected figures. Tests of significance of differences between sexes, or between sibships, are carried out in the usual manner, using the mean squares based upon the expected numbers and sums for the effects being tested, and comparing with the mean square for individual variability within subclasses, from the primary analysis (Table VII).

For body length in the Round Lake stock, which has been worked out in detail in Table VII as an example, highly significant differences exist between the sexes, and at the same time, between some of the sibships. Differences between sexes in the remaining linear measurements from the Round Lake mice were highly significant in femur length, mandible length, and condyle-zygoma length, and significant in condyle-premaxilla length (Table VIII). Differences between sexes are not significant in tail length, foot length, ear length, and bullar width.

Between sibships in the Round Lake stock, differences were highly significant in all measurements except in ear length, in which differences were at the level termed significant.

Interaction between sex and sibship is appreciable only in two lengths, in tail length, in which it was highly significant, and in ear length, in which it was significant. Interaction between sex and sibship indicates that the sex difference varies significantly among the various sibships. In the tail-length data, for example, the females are larger than the males in several sibships but smaller in others, and this variation is significantly greater than the variability within groups of like-sexed sibs.

Body-length data throughout the nine stocks, when examined as to variability (Table IX), show that highly significant differences between sibships are present in six of the stocks, a significant difference exists in the Daleville stock, and there are no significant differences between sibships in the Santa Rosa Island and the Panama City stocks. A highly significant difference between sexes is present in six stocks, with no significant differences in the stocks from Destin, Panama City, and Daleville.

In the foot-length data (Table IX), highly significant differences between sibships are present in the Seminole Hills, Round Lake, and Sills stocks; significant differences in the Crystal Lake and Marianna stocks; and no significant differences in the three beach stocks from Santa Rosa Island, Destin, and Panama City or in the inland stock from Daleville.

In summary, in body and skeletal measurements and in color measurements significant differences between sibships exist in some stocks and not in others and also in some measurements of mice of a particular stock and not in other measurements of the same animals. Differences between sexes are absent in the color measurements and in the foot-length measurements,

TABLE VIII

ANALYSIS OF VARIANCE FOR BODY AND SKELETON MEASUREMENTS (EXCEPT BODY LENGTH) IN *Peromyscus polionotus albifrons* FROM ROUND LAKE

Source of Variability	Tail Length		Foot Length		Ear Length		Femur Length	
	Degrees of Freedom	Mean Square	Degrees of Freedom	Mean Square	Degrees of Freedom	Mean Square	Degrees of Freedom	Mean Square
Between Sexes	1	14.54	1	.06	1	.38	1	2.71**
Between Sibships	7	59.55**	7	1.21**	7	.48*	7	.31**
Interaction (Sex × Sibship)	7	24.93**	7	.18	7	.60*	7	.06
Error	62	4.09	62	.13	62	.21	58	.06

Source of Variability	Mandible Length		Condyle-premaxilla Length		Condyle-zygoma Length		Bullar Width	
	Degrees of Freedom	Mean Square	Degrees of Freedom	Mean Square	Degrees of Freedom	Mean Square	Degrees of Freedom	Mean Square
Between Sexes	1	.58**	1	59*	1	1.02**	1	.00
Between Sibships	7	.22**	7	.75**	7	.23**	7	.11**
Interaction (Sex × Sibship)	7	.06	7	.06	7	.08	7	.03
Error	60	.06	59	.11	60	.07	60	.02

* Significant. ** Highly significant.

TABLE IX

ANALYSIS OF VARIANCE FOR BODY LENGTH AND FOOT LENGTH IN *Peromyscus polionotus*

Stock	Source of Variability	Degrees of Freedom	Mean Squares	
			Body Length	Hind-Foot Length
Santa Rosa Island	Between Sexes	1	118.7**	.86
	Between Sibships	4	15.3	1.66
	Interaction	4	4.7	1.90
	Error	18	6.4	.73
Destin	Between Sexes	1	1.2	.40
	Between Sibships	2	22.1**	.71
	Interaction	2	.03	1.20*
	Error	18	2.8	.29
Panama City	Between Sexes	1	1.5	.10
	Between Sibships	1	11.1	.09
	Interaction	1	5.0	.39
	Error	5	3.6	.24
Seminole Hills	Between Sexes	1	94.3**	.34
	Between Sibships	6	86.9**	1.05**
	Interaction	6	6.0	.16
	Error	49	4.0	.31
Crystal Lake	Between Sexes	1	199.3**	.01
	Between Sibships	8	21.7**	.93*
	Interaction	8	4.8	.18
	Error	40	4.0	.24
Round Lake	Between Sexes	1	97.3**	.06
	Between Sibships	7	15.4**	1.21**
	Interaction	7	2.5	.18
	Error	62	4.8	.13
Marianna	Between Sexes	1	185.1**	.12
	Between Sibships	9	27.1**	.57*
	Interaction	9	42.6**	.16
	Error	70	5.5	.23
Sills	Between Sexes	1	40.1**	.06
	Between Sibships	5	51.8**	1.98**
	Interaction	5	2.6	.02
	Error	34	2.7	.12
Daleville	Between Sexes	1	6.8	.86
	Between Sibships	3	16.7*	.76
	Interaction	3	6.6	.24
	Error	15	3.2	.48

* Significant.
** Highly significant.

but a sex difference is apparent in body length in most stocks, and in certain skeletal measurements examined only in the Round Lake mice.

Meaning of Differences Between Sibships

It is important to an understanding of the present problem to consider the reasons why differences between sibships may exist in some measurements and not in others. Differences arising from sampling procedures will be discounted for the present, since allowance for them is made in the analysis of variance.

Significant differences between sibships are assumed to be largely hereditary. Nonhereditary factors causing differences between sibships may also be present (Wright, 1920; Lush, 1945), although an attempt has been made to minimize their effect by keeping the laboratory conditions under which the mice were reared as uniform as possible for all mated pairs of mice.

When significant differences do exist between sibships, they are assumed to be the result, at least in large part, of hereditary differences between the pairs of parents. When significant differences cannot be demonstrated with the data at hand, however, either the hereditary differences between the sets of parents are nonexistent, or the differences are too small to be revealed by the sample at hand.

Hereditary differences between sets of parents will vary in relation to the heterozygosity of the population from which the parents are drawn. Significant differences between sibships indicate some heterozygosity. Lack of significant differences between sibships indicates that it is not possible, with the data at hand, to demonstrate that the stock is not homogeneous for the hereditary factors governing the character studied.

For pelage-color characteristics there is at least a possibility that a number of stocks are relatively homozygous for genes governing pelage color, whereas in other stocks, or in other color characteristics in the same stocks, some amount of heterozygosity is apparent (Table V). In the Santa Rosa Island and the Panama City stocks there is no evidence that the animals are not homozygous for the genes governing each of the three colors read, dorsal and side. In the Marianna stock the animals may be homozygous for the genes governing dorsal green and blue-violet, but they are quite certainly heterozygous for at least some of the genes governing all other colors read.

Body-length and foot-length data (Table IX) agree with the color measurements in showing that the genetic variability in a stock of mice may vary according to the characteristic being studied.

Differences Due to Age

Age differences are difficult to analyze exactly from the data at hand. When measurements from two-year-old animals are available, only the roughest of comparisons may be made. In the Santa Rosa Island stock eighteen two-year-old animals were raised. Comparison of the two-year animals with twenty-eight one-year-olds, segregated according to sex but not according to sibship, using body and bone measurements, shows that in some measurements, especially body and tail length, the older animals are the larger (Table X). Because of the small numbers, and the impossibility of including the sibship factor, no statistical analysis of these data has been attempted.

In instances in which color measurements are available for mice of dif-

TABLE X

Body and Skeleton Measurements in Millimeters of *Peromyscus polionotus leucocephalus* from Santa Rosa Island

Sex	Age Class		Body Length	Tail Length	Foot Length	Ear Length
Female	One Year	Number	18	18	18	18
		Mean	79.39	54.83	18.77	15.27
	Two Year	Number	5	5	5	5
		Mean	81.40	59.60	19.12	15.74
Male	One Year	Number	10	10	10	10
		Mean	75.40	52.80	18.83	15.05
	Two Year	Number	13	13	13	12
		Mean	76.62	54.38	18.86	15.58

Sex	Age Class		Femur Length	Mandible Length	Condyle-premaxilla Length	Condyle-zygoma Length	Bullar Width
Female	One Year	Number	18	18	18	18	18
		Mean	15.27	14.63	22.56	16.72	10.46
	Two Year	Number	3	3	3	3	3
		Mean	15.60	14.90	22.63	16.73	10.53
Male	One Year	Number	10	9	8	9	9
		Mean	14.54	14.36	22.05	16.46	10.33
	Two Year	Number	9	9	9	9	9
		Mean	14.55	14.51	22.22	16.42	10.33

ferent ages, there seemed to be no appreciable differences between older and younger adult animals. Very young mice, of course, are paler, not having attained the adult pelage. Such subadult animals were excluded from the analyses. The averages of color readings of adult Santa Rosa Island animals (Table XI) are based on so few animals as to be highly variable, but show no general trend for age or sex.

TABLE XI

Pelage Color Measurements in *Peromyscus polionotus leucocephalus* from Santa Rosa Island, in per cent

Sex	Age Class	Number of Animals	Dorsal Readings			Side Readings		
			Red	Green	Blue-violet	Red	Green	Blue-violet
Male	One Year	6	18.3	15.0	13.2	25.5	21.2	18.0
	Two Year	3	19.0	14.7	12.7	30.0	25.3	22.7
Female	One Year	2	23.3	14.0	13.0	20.0	19.3	15.7
	Two Year	9	19.6	16.0	14.6	27.6	22.8	20.0

Analysis of the Structure of the Sample

The sample consists of the wild mice trapped from the wild population, as explained above when describing the mean. The characteristics studied are measured in the offspring of pairs of the wild-caught mice, and each sibship may be considered a sample of the combined genetic potentialities of the two parents. Because of the mechanics of this sampling system, each pair must be considered a single item in the sample of the wild population even though each pair consists of two individuals. The number of pairs of parents (or number of sibships) thus becomes the number of items in the sample.

There are two sources of the differences between sibship means within one stock: first, the real differences between the true sibship means, coming from a real difference in genetic make-up and also from any nonhereditary differences associated with sibships, and second, apparent differences between sibship means, arising from the sampling method (breeding) employed in determining the means. If no true differences were present between sibships, apparent differences would still be expected, just as in any case of sampling a homogeneous population of normal variability.

The best sample of any population will be that in which the part of the variability due to the sampling method used is at a minimum. In some types of sampling, as in the present one, a change in the details of the sampling process will decrease that part of the variability within the sample due to sampling method. The less the effect of this type of variability, the more true will be the sample's representation of the parent population.

Similar problems of sampling and subsampling in other fields have been attacked by a number of workers, including Immer (1932), Yates and Zacopanay (1935), and Cochran (1938), using the same general methods outlined by Fisher (1944) and by Snedecor (1940, 1946). Snedecor's (1940) notation is retained in the present discussion. While these original papers deal with problems in field crops, the following discussion is in terms of the problem of the present study.

The two sources of variance which contribute to differences between sibships may conveniently be identified by symbols, as "A," the true variance between sibship means caused by the real differences among sibships, and "B," the variance between individual offspring within sibships, resulting from the sampling used to determine sibship means.

Variance A is that due to hereditary differences between sibships and to such nonhereditary differences as may remain associated with the individual pairs of parent mice. The nonhereditary differences have been reduced as much as possible by attempting to provide a uniform laboratory environment and will, henceforth, be ignored in this discussion. Variance A cannot be determined directly by experiment, and may only be estimated through a sampling procedure.

Variance B is that due to the variability between the members of sibships and arises from several sources. Genetic differences arising from segregation and recombination of the germinal material which occur between members of the same sibship, differences due to environmental effects, and errors made in making measurements will show their effect here.

In the wild population the means of sibships have a distribution with a variance A, the size of this variance being determined by the hereditary make-up of the parents. But when an investigator samples the wild population by determining the means of a number of sibships, his estimate of the variance between means must be larger than the true variance, A, because of the use of sampling methods. The added variance due to the effect of sampling is $\frac{B}{k}$, when samples of size k and variance B are used to determine each sibship mean. (In any population of individuals with variance B, means of random subsamples composed of k individuals each, will be distributed with a variance $\frac{B}{k}$.) The two portions of the estimated variance between sibship means may be added if it is assumed that they are independent (Fisher, 1944; Snedecor, 1946). Thus the estimate of the variance of the distribution of sibship means becomes $A + \frac{B}{k}$, when k offspring per sibship are used, and individuals vary within sibships with variance B.

If there are unequal numbers of offspring in the sibships, an average number must be used. According to Cochran (1938) and Snedecor (1946) this average number represented by k_0, must be obtained by the formula

$$k_0 = \frac{1}{n-1}\left(\Sigma k - \frac{\Sigma k^2}{\Sigma k}\right),$$

where n is the number of subsamples and k is the number of items in any particular subsample. This formula gives a value equal to the arithmetic mean of the numbers in the various subsamples when those numbers are all identical. In other instances, k_0 is less than the arithmetic means. Values of k_0 for the color data are given in Table III.

In order to obtain an estimate of the variance of individual items, based upon the distribution of sibship means, such as might be used in an analysis of variance table, the quantity $A + \frac{B}{k}$, the estimated variance of the distribution of sibship means, must be multiplied by the average number of offspring per sibship (designated as k in the following general discussion, although in actual computation the value k_0 must be used.) Table XII is a symbolic representation of the different estimates of variance, in the form of an analysis of variance table. The total sum of squares is obtained by adding its two component parts, derived from within-sibship and between-sibship variation.

TABLE XII

SYMBOLIC REPRESENTATION OF VARIANCE ESTIMATES, WHEN THE SAMPLE IS DRAWN FROM THE WILD POPULATION BY BREEDING CAPTURED MICE, WITH n SIBSHIPS, EACH CONTAINING k OFFSPRING

Source of Variability	Degrees of Freedom	Sums of Squares	Mean Square
Total	$nk-1$	$(nk-n)\ B+(n-1)\ (kA+B)$	
Between Sibships	$n-1$	$(n-1)\ (kA+B)$	$kA+B$
Within Sibships	$nk-n$	$(nk-n)\ B$	B

$$F=\frac{kA+B}{B}=1+\frac{kA}{B}$$

From Table XII the nature of the variance ratio, or "F" value, may be seen. If there are no true differences between sibships, the F value,

$$\frac{kA+B}{B},$$

will be approximately unity, varying only because of the sampling effect. True differences between sibships will increase the F ratio, although true differences may be present and yet too small to be reliably estimated from the sample used. Estimates of values of variances A and B remaining the same, the F ratio will increase as the number of offspring per sibship is increased.

The value of variance A may be estimated. Since the analysis of variance table contains two estimates of variance B, the estimate of A is derived by subtracting the value of B (mean square for individuals within sibships) from the value of $kA+B$ (mean square for between-sibship means) and dividing by k (the average number of individuals per sibship) (Snedecor, 1946).

$$A=\frac{(kA+B)-B}{k}.$$

Values of estimates of variances A and B in the six color measurements for the different stocks are given in Table XIII.

As was pointed out by Henry, Down, and Baten (1942), this method of analysis of the sources of error at times gives impossible numerical results. Logically, no variance can have a negative value, yet in their work, as in the present problem, negative values of A appear. In such cases they use a relationship identical with that of assuming a value of zero for A. In the present study, wherever a negative value of A was found, as happened in several cases, A was assigned the indefinite value of "small."

A and B are variances, and mere estimates of their values are subject to variation in themselves. Since any estimate of A must be computed from two mean squares, each containing an independent estimate of B, it would seem that when the true value of A is small compared with that of B, an apparent negative estimate of A might result solely from the chance variation of the two estimates of B. Snedecor (1946) shows estimates of variance

A, which include negative values, obtained in sampling a homogeneous population.

One reason for computing variances A and B lies in the use of these quantities to reconstruct an estimate of the experimental error under various apportionings of k, the number of animals per sibship, and n, the number of sibships per stock. As shown above, the mean square due to differences between sibship means is $kA+B$. To convert to the estimated variance of sibship means (on the sibship basis), $kA+B$ is divided by k, the number of individuals per sibship, giving $A+\frac{B}{k}$ as the variance of sibship means. The variance of the stock mean, estimated from the distribution of the sample of sibships used, results from the division of the variance of sibship means by n, the number of sibships in the stock. The result,

$$\frac{A}{n}+\frac{B}{nk} \qquad (1)$$

TABLE XIII

ESTIMATES OF VARIANCE IN PELAGE COLOR MEASUREMENTS OF *Peromyscus polionotus*

If the actual laboratory population were divided into n sibships, each with k offspring, the optimum (lowest) value of the variance of the mean (V_m) could be expected.

Color	Stock	Estimates of Variance		Optimum Values			Present V_m
		A	B	n	k	V_m	
Dorsal Red	Santa Rosa Island	.880	3.54	6.0	2.8	.355	.693
	Destin	.568	.82	12.7	1.7	.083	.182
	Panama City	Small	1.87	1.0	30.0	.062#	.160
	Seminole Hills	.105	1.08	17.5	4.5	.020	.036
	Crystal Lake	Small	2.68	1.0	133.0	.020#	.094
	Round Lake	.159	1.38	14.9	4.2	.033	.057
	Marianna	.362	1.63	26.6	3.0	.034	.173
	Sills	.041	1.56	6.6	8.7	.033	.056
	Daleville	.609	1.65	11.1	2.3	.119	.261
Dorsal Green	Santa Rosa Island	.811	5.18	5.2	3.6	.435	.823
	Destin	.115	.31	10.9	2.3	.023	.046
	Panama City	Small	1.75	1.0	30.0	.058#	.111
	Seminole Hills	.088	.53	20.8	3.5	.012	.023
	Crystal Lake	Small	2.22	1.0	133.0	.017#	.064
	Round Lake	.364	.93	21.6	2.3	.036	.085
	Marianna	.065	1.10	17.0	5.8	.015	.066
	Sills	.072	.75	10.8	4.6	.022	.035
	Daleville	.357	.72	12.0	2.0	.060	.137
Dorsal Blue-violet	Santa Rosa Island	.677	2.61	6.1	2.8	.266	.522
	Destin	.347	.79	11.4	2.1	.063	.131
	Panama City	Small	1.45	1.0	30.0	.048#	.073
	Seminole Hills	.502	.67	31.4	1.6	.029	.077
	Crystal Lake	Small	2.71	1.0	133.0	.020#	.053
	Round Lake	.489	.74	24.6	1.7	.037	.104
	Marianna	.044	.82	16.4	6.1	.011	.048
	Sills	Small	.68	1.0	69.0	.010#	.018
	Daleville	.379	1.03	11.1	2.3	.074	.162

# Assuming variance A to be negligible where the numerical estimate of this variance is negative.

TABLE XIII—(*Cont.*)

Color	Stock	Estimates of Variance		Optimum Values			Present V_m
		A	B	n	k	V_m	
Side Red	Santa Rosa Island	2.039	23.36	4.2	4.8	1.619	3.062
	Destin	.475	3.01	8.4	3.6	.156	.310
	Panama City	.006	6.57	1.0	30.0	.225	.695
	Seminole Hills	.472	4.16	18.4	4.2	.079	.149
	Crystal Lake	2.190	12.17	25.3	3.3	.201	.843
	Round Lake	.888	4.54	17.7	3.2	.130	.253
	Marianna	1.472	3.82	31.2	2.2	.101	.583
	Sills	.646	1.72	16.5	2.3	.084	.161
	Daleville	Small	3.10	1.0	46.0	.067#	.070
Side Green	Santa Rosa Island	Small	23.64	1.0	27.0	.876#	1.582
	Destin	Small	1.97	1.0	45.0	.044#	.087
	Panama City	Small	3.32	1.0	30.0	.111#	.321
	Seminole Hills	.325	2.43	37.3	1.0	.070	.093
	Crystal Lake	1.365	5.49	27.9	2.8	.118	.443
	Round Lake	.988	2.13	22.6	2.1	.089	.223
	Marianna	1.279	2.31	34.1	1.9	.073	.464
	Sills	.084	.98	10.4	4.8	.028	.045
	Daleville	.304	.93	10.7	2.5	.063	.137
Side Blue-violet	Santa Rosa Island	1.100	17.68	3.8	5.7	1.116	2.139
	Destin	Small	1.60	1.0	45.0	.036	.089
	Panama City	.413	2.49	5.8	3.5	.193	.400
	Seminole Hills	.430	1.68	23.8	2.8	.043	.090
	Crystal Lake	.710	7.16	20.8	4.5	.111	.400
	Round Lake	1.183	1.34	26.2	1.5	.079	.242
	Marianna	1.176	2.36	41.6	1.2	.076	.436
	Sills	Small	.82	1.0	69.0	.012	.014
	Daleville	.149	1.03	8.4	3.7	.051	.105

# Assuming variance A to be negligible where the numerical estimate of this variance is negative.

will be an estimate of the variance of the stock mean, and its square root will be the standard deviation of the stock mean (standard error).

Most Efficient Sample

Now, since A and B are estimates of quantities constant in one measurement for any stock, the question remains as to what choice of number of sibships (n) or number of offspring per sibship (k) will produce the smallest variance of the stock mean, that is to say, the variance least increased by the influence of the sampling method used.

Any increase in n will cause a proportional decrease in the variance of the stock mean, but the reduction caused by increasing k will depend upon the relative size of variances A and B (see formula 1, above). When B is small compared with A, a change in k, the number of animals per sibship, will produce very little change. If A is smaller than B, however, a change in either n or k will produce about equal effect. Thus, if there were no other limitation, increasing the number of sibships (n) would always reduce the variance of the stock mean and also the square root of that variance, the standard error. Increasing the number of animals per sibship (k) would never be more effective and might have little effect.

For reducing the estimated variance of the stock mean, the advantages of increasing n are so great that such a move would seem always desirable, except when the relative cost of gathering the data is too great. This relationship has been discussed by Fisher (1944) and Snedecor (1946) and in its application to sampling wheat fields by Cochran (1938) and King and Jebe (1940). In the present problem, it seems clear that the best sample will consist of the largest possible number of sibships, each containing, if necessary, a minimum number of offspring.

In applying this method of analysis of the composition of the variance of the stock means to the present study, one modification has been made. This change is simply an extension of the general theory to this special case where the data relate to sibships of mice. In such a problem as this, the efficiency of the sampling method may be estimated, involving directly the cost of using a greater or lesser number of sibships. Efficiency may here be understood as the relationship between the information at hand and that information which might have been collected by an altered method of sampling or analysis, using the same effort or materials.

In studies involving laboratory breeding of mice, there is ordinarily a limit on the effort which can be devoted to any experiment. This limit is expressed in terms of the equipment, space, and services possible to devote to the study, in other words, the number of mice which can be cared for. The question of efficiency of the experiment is thus resolved into making the best use of a certain number of animals. Each sibship has two parents which take up laboratory space, and which do not contribute directly to the data. With a fixed laboratory-population number, the larger the number of sibships raised, the larger will be the number of parents, and the smaller will be the proportion of the laboratory population useful for measurements. In the most extreme case each sibship would contain only one offspring, and two-thirds of the laboratory animals would be parents. This would be the condition of the maximum number of sibships and the minimum number of offspring in each sibship.

Since laboratory space is limited, there will be one division of the laboratory population into parents and offspring which will give the most efficient results in terms of the variance of the stock mean. If L is the numerical limit of the laboratory population, n the number of sibships, and k the number of individuals per sibship, then n cannot exceed one-third of L and

$$L = 2n + nk$$

$$k = \frac{L}{n} - 2. \qquad (2)$$

Naturally, as n increases, k decreases. Substituting this value of k in relationship 1, the variance of the stock mean becomes

$$\frac{A}{n}+\frac{B}{n\left(\frac{L}{n}-2\right)},$$

or

$$\frac{A}{n}+\frac{B}{L-2n}.$$

In this expression, the values of A and B are constants, related to the variability of the population. The size of the laboratory population (L) is also a constant. The variance of the stock mean will vary with the size of n, and will reach a minimum value when one certain value of n is used. This value of n may be found either by trial-and-error computation or by obtaining the first derivative of the variance with respect to n and equating it to zero. Under these conditions

$$n=L\left[\frac{\sqrt{2AB}-2A}{2B-4A}\right], \tag{3}$$

and, from (2),

$$k=\sqrt{\frac{2B}{A}}.$$

Since estimates of A and B may be computed from the ordinary analysis of variance, the value of their ratio may be used in computing a value of k. For any particular ratio of A to B, k appears to be constant regardless of L (the laboratory population). The value of n (the number of sibships) will vary with L (see formula 2).

Having now the value of n and k which would have resulted in the smallest value for the variance of the stock mean had the actual laboratory population been so divided (Table XIII), an estimate of this smallest value of the variance may be computed, using the values of A and B already obtained. The percentage difference between this best possible value of the variance and the actual value may be used as an estimate of the loss in efficiency from not using the best distribution of animals among sibships and offspring.

Values for the potential gain in efficiency are shown in Table XIV. These figures may also be viewed as measuring potential gain in information from the same effort expended (Fisher, 1944). The general low efficiency of the present samples may arise from any or all of three causes:

1. The laboratory population is not divided into the optimum number of sibships and offspring within sibships.
2. The sibships have different numbers of offspring, and the effective average number (k_o) is less than the numerical average number of offspring per sibship.
3. The effective number of sibships (Table III) is small, due to the small

TABLE XIV

Percentage Gain in Information in Pelage Color Measurements of *Peromyscus polionotus* Which Would Result Were the Actual Laboratory Population To Be Divided Into Optimum Numbers of Sibships and Offspring

Stock	Dorsal Readings			Side-Stripe Readings		
	Red	Green	Blue-violet	Red	Green	Blue-violet
Santa Rosa Island	49	47	49	47	45†	48
Destin	55	51	52	50	50†	60†
Panama City	61†	47†	34†	68	66†	52
Seminole Hills	46	49	62	47	25	52
Crystal Lake	78†	74†	62†	76	27	72
Round Lake	43	58	64	48	60	67
Marianna	80	77	77	83	84	83
Sills	40	38	46†	48	38	12†
Daleville	54	56	54	4†	54	52

† Assuming variance *A* to be negligible where the numerical estimate of this variance is negative.

number of wild parents comprising the ancestry of the laboratory-bred animals.

The actual laboratory population in the Marianna stock, for example, is made up of 103 offspring in fifteen sibships, or at least 133 animals in all. The laboratory-bred mice are all descended from seven wild-caught animals (3.5 effective sibships), and the effective average number of offspring per sibship is 6.7 (Table III), so that the effective laboratory population is approximately thirty-one, instead of the actual 133 animals. The other stocks are reduced in effectiveness in a similar manner.

Greater efficiency in sampling technique is desirable, especially in view of the potential improvement which Table XIV shows to be possible. Probably absolute efficiency is impossible of attainment, especially where a number of characteristics are studied. Certain suggestions, based upon the previous discussion and upon the data in Tables XIII and XIV, may be offered for general improvement of sampling.

The optimum number of offspring per sibship is dependent upon the ratio between the inherited variability and the noninherited variability. This ratio is a constant within a stock for any particular characteristic, but may vary widely within a stock between one characteristic and another (Table XIII). The best number of offspring, as judged from these data, seems to be most often between two and five animals per sibship, in situations wherever hereditary variability is important. On the other hand, where hereditary variability is of little importance, greatest efficiency results from the rearing of many offspring in a few sibships.

The optimum number of sibships to be used also depends upon the ratio of inherited to noninherited variability. Since the number of sibships is the number of items in the sample, the desirable number of sibships further depends upon the magnitude of the variance of the mean (or its square root, the standard error) which the investigator will accept as indicating an adequate sample. If a desirable value for the standard error can be stated, then a simple computation will estimate the necessary number of sibships. The square of the desirable standard error may be substituted as the variance of the mean (V_m) in

$$V_m = \frac{A}{n} + \frac{B}{nk} \tag{1}$$

and the equation solved for n, where k is already determined by the ratio of hereditary to nonhereditary variability. Such a computation assumes that the variances arising from hereditary and nonhereditary factors have been estimated.

In the sampling of a population, a decision on sampling method has necessarily to be made before the data are collected and, therefore, before there is any indication as to the relation between hereditary and nonhereditary variability. The number of sibships is often limited by the number of fertile wild mice captured during the field work. Later, while rearing the laboratory-bred offspring, it is possible to alter the number of offspring per sibship, but not to increase the number of wild-caught parents except by further field-collecting. This is time-consuming, expensive, and often impossible. Practically, the number of wild parents captured in the field limits the effective number of sibships.

The number of sibships is so important in the type of sampling here discussed that an investigator would better plan to raise two to five offspring per sibship and to devote all additional effort in the laboratory and in the field to increasing the number of sibships. Such a sampling plan as this is calculated to be most efficient in situations where hereditary variability is important. To increase the number of offspring per sibship at the expense of the number of sibships is to risk finding that the data do not adequately sample the population in which hereditary variability is important.

The preceding discussion relates only to the advantages of increased numbers of sibships in reducing the standard error. A different relationship holds where the interest lies chiefly in comparing the importance of hereditary and nonhereditary variability within a particular population.

Inherited Proportion of the Variability

The relative importance of real differences between sibships in causing variability may be estimated from the ratio of that portion of the variance due to these differences (variance A) to the total variance ($A + B$). Since true differences between sibships arise from both hereditary factors and nonhereditary factors associated with the individual sibships, such a ratio of variances is an estimate of the largest portion of the variability ascribable to hereditary factors. The true proportion of variability inherited will be somewhat smaller, as it depends upon the here unknown importance of these nonhereditary differences. More detailed experiments, such as those of Wright (1920), Gowen (1934), Lush, Hetzer, and Culbertson (1934), Bywaters (1937), and Powers (1942), would better determine the importance of nonhereditary differences between sibships.

Fisher (1944) showed the identity of the ratio of variances mentioned above with the intraclass correlation coefficient,

$$\frac{A}{A+B},$$

and he said that "the intraclass correlation will be merely the fraction of the total variance due to that cause which observations in the same family have in common." Gini (1940) showed the relationship between the intraclass correlation coefficient and a ratio of variances very similar to the above when large samples are used.

In the present study, estimates of variances A and B were used to compute r_I according to Fisher's formula. Values of this coefficient for the various color measurements are given in Table XV. The F test, applied earlier in examination of sibship differences by analysis of variance (Table V), is a test of the significance of the difference from zero of the computed estimates of intraclass correlation. Where differences are shown to exist between sibships, the value of r_I is known to be greater than zero.

TABLE XV

INTRACLASS CORRELATION COEFFICIENTS FOR PELAGE COLOR MEASUREMENTS IN *Peromyscus polionotus*

A negative numerical estimate is designated "S."

Stock	Dorsal Colors			Side Colors		
	Red	Green	Blue-violet	Red	Green	Blue-violet
Santa Rosa Island	.20	.14	.21	.08	S	.06
Destin	.41*	.28	.31*	.14	S	S
Panama City	S	S	S	.002	S	.14
Seminole Hills	.08	.14*	.43**	.10	.12	.20**
Crystal Lake	S	S	S	.15*	.20*	.09
Round Lake	.10	.28**	.40**	.16*	.32**	.47**
Marianna	.18**	.05	.05	.28**	.36**	.33**
Sills	.02	.08	S	.27*	.08	S
Daleville	.27	.33*	.27	S	.24	.13

* Difference from zero significant.
** Difference from zero highly significant.

Where the sibship mean is the arithmetic mean of the values for the two parents, and mating is at random, the variance between sibships should not exceed half the total variance of the wild population. This is true because the variance of a series of means of two-item subsamples, formed at random, will be one-half of the variance of the population of individuals from which the subsamples are drawn. Nonrandom mating and nonhereditary differences between sibships will act to increase the value of r_I, but factors increasing the variability within sibships, such as environmental effects or random errors in measurement, will tend to decrease the value of r_I.

In interpreting the data in Table XV, it should be borne in mind that values of r_I should not be expected to be much, if any, over .50. The confidence limits of the estimates of intraclass correlation in the present study are rather broad, as indicated by the size of some of the values not significantly different from zero. Detailed study would require more adequate data.

For all the limitations of the data, it is apparent that some differences do exist between stocks in the proportion of the variability in color measurements which is inherited. Sumner's (1929) finding of greater variability in the "intergrade populations" may be confirmed by the greater genetic variability in the Round Lake and Marianna stocks, though his impression of greater variability in all stocks of the subspecies *albifrons* is not supported by the present data.

A low value of the intraclass correlation coefficient in Table XV indicates a possible approach to genetic homogeneity, since these coefficients are estimates of the proportion of variability which is inherited. In the color data over half the estimates of the maximum importance of hereditary variability are 10 per cent or less. In two-thirds of the instances the value is not significantly different from zero, and half of the significant differences are confined to the Round Lake and Marianna stocks. This signifies that, in a large proportion of the different combinations of genes regulating pelage color, the various populations of mice may be approaching genetic homogeneity.

The body-length and foot-length data as opposed to the color data, indicate that a generally greater proportion of the variability is due to hereditary factors. While the intraclass correlation coefficient was not computed for these measurements, Table IX shows that in body length significant differences exist between sibships (therefore some genetic differences are probably present) in all stocks except the two from Santa Rosa Island and Panama City.

In the foot-length measurements, significant differences between sibships appear in all but one (Daleville) of the six inland stocks but in none of the three beach stocks.

Standard Deviation

The usefulness of the standard deviation in comparing populations (Hubbs and Perlmutter, 1942; Dice, 1944, 1944*a*) makes it a desirable estimate of variability. Yet a standard deviation derived from the distribution of sibship means would have little meaning when referring to individuals within a wild population. The desired standard deviation is that based upon the total variance, as estimated by the sample.

The total variance of the population from which the samples are drawn is the sum of variances A and B (Snedecor, 1946). The estimate of the total variance in the sample is not, however, an accurate estimate of the total variance in the wild population. It should be recalled that the sample is here drawn according to sibship.

As given in Table XII, the total sum of squares in the sample is

$$(nk-n)\ B+(n-1)\ (kA+B).$$

When divided by the corresponding degrees of freedom $(nk-1)$, this equals, as an estimate of total variance

$$A\left(\frac{nk-k}{nk-1}\right)+B.$$

The estimate of total variance differs from the true value $(A+B)$ not only because of sampling variability in estimating variances A and B, but also because variance A is represented by only a fraction of its estimated value. This fraction, $\frac{nk-k}{nk-1}$, has a value of zero when one sibship is used, and no estimate of variance A is then available. With two sibships, the value is somewhat over one-half; with three, over two-thirds; so that, if a reasonable number of sibships are sampled, the value of the fraction approaches unity. Thus, the error, which is due to inadequate sampling of hereditary variability, is reduced by the use of more sibships.

It is of some value to estimate the possible error from computing the standard deviation from laboratory-bred animals without regard to sibship, as has been done in a number of past studies. The greatest error would arise when a single sibship was used and no value of variance A represented. In such a case, hereditary variability of the stock would not be sampled. If the single sibship used furnished a good estimate of variance B, the estimated total variance (simply variance B in this case) would be at least half the sum of $A+B$ (since variance A is not expected to be over half this sum). The standard deviation, therefore, would be, on the average, at least seven-tenths of the value of a better estimate. This does not include any aspect of possible variation in estimating variance B due to a small sample.

In the event that there are no hereditary differences between sibships, the value of variance A is zero, and any estimate of total variance simply estimates variance B, and is based upon variability between individuals. Judging by the present data, there may be a number of stocks where the color data provided by a few sibships would lead to the same conclusions as when more sibships are used.

Thus the standard deviation as computed in past studies may be a reasonably dependable estimate of the true value when a number of sibships are used, but when a poor sample of the hereditary variability of the wild population is used, the computed value may be underestimated by a considerable margin. The importance of the error depends upon the proportion of the variability which is inherited, and this factor cannot be judged from a poor sample.

TABLE XVI

Standard Deviations for Color Measurements in *Peromyscus polionotus* Computed by Two Methods

1. Computed from all laboratory-bred offspring as one population, without regard to sibship. 2. Computed as the square root of the sum of variances *A* and *B*.

Stock	Dorsal Readings					
	Red		Green		Blue-violet	
	1	2	1	2	1	2
Santa Rosa Island	2.02	2.10	2.38	2.45	1.74	1.81
Destin	1.10	1.18	.62	.65	1.01	1.07
Panama City	1.33	1.37	1.25	1.32	1.12	1.20
Seminole Hills	1.08	1.09	.78	.82	1.05	1.08
Crystal Lake	1.62	1.64	1.44	1.49	1.59	1.65
Round Lake	1.23	1.24	1.11	1.14	1.08	1.11
Marianna	1.40	1.41	1.08	1.08	.93	.93
Sills	1.26	1.27	.90	.90	.82	.83
Daleville	1.44	1.50	1.01	1.04	1.16	1.18

Stock	Side Readings					
	Red		Green		Blue-violet	
	1	2	1	2	1	2
Santa Rosa Island	4.96	5.04	4.75	4.86	4.28	4.33
Destin	1.84	1.86	1.38	1.40	1.26	1.27
Panama City	2.56	2.56	1.96	1.82	1.68	1.70
Seminole Hills	2.12	2.15	1.65	1.66	1.43	1.45
Crystal Lake	3.76	3.79	2.60	2.62	2.80	2.81
Round Lake	2.30	2.33	1.72	1.77	1.53	1.59
Marianna	2.28	2.30	1.87	1.90	1.85	1.88
Sills	1.50	1.54	1.02	1.03	.87	.90
Daleville	1.62	1.76	1.09	1.11	1.08	1.09

Table XVI gives the values of standard deviations for all color measurements as computed, (1) from the laboratory-bred animals as one population, without regard to sibship, and (2) as the square root of the sum of estimates of variances *A* and *B*. Wherever the numerical estimate of variance *A* is negative, variance *B* was used alone, assuming a negligible value for *A*. Examination of Table XVI will show that in these color data the difference between the two estimates is small. In these data, therefore, the use of a standard deviation calculated from the laboratory population without regard to sibships would not lead to truly erroneous conclusions.

Standard Error

Since the sampling is done by sibships, a standard error for comparing stock means must be based upon sibship means. This standard error is the square root of the estimated variance of the stock mean, which is obtained by dividing the estimated variance based upon sibship means by the product of the number of sibships and the average number of offspring per sibship (formula 1).

If the interest of the study lies solely in comparing populations according to differences between means of samples, a second method of computing the standard error is available. The estimates obtained by this method are more crude than by the first computation but have the advantage of being less laborious to calculate. In the case of the body and bone measurements, this second method avoids the awkwardness of disproportionate subclass numbers.

Since the fundamental sampling unit is the sibship mean, in this second method each sibship mean is used as a single value, no matter how many or how few mice may have been in that sibship. The average squared deviation from the stock mean is computed by the usual method and divided by the number of sibships to yield an estimate of the stock variance. The square root of this last quantity is the standard error.

Fundamentally, the effect of numbers of mice in the sibship is to determine the sibship means with greater precision, and only indirectly to affect the stock mean. Although this second method does not allow, in computation, for varying numbers within the sibships, all effect of greater number within sibships is not lost. Other things being equal, as numbers within the sibship increase, the variability between sibship means will reflect less and less the variability between individuals within sibships, and the residual variability will represent more truly the real variability between sibships. This second method is not ''throwing away data'' but only using the data to derive a somewhat less refined estimate of a statistical characteristic, which is, in this study, at best only roughly determined.

The actual difference between the two methods is one of weighting the contribution of each sibship to the variance of the mean, in accordance with the number of animals in that sibship. In the first method, the contribution of sibships larger than the average is magnified and that of the smaller sibships is made less important. By the second method, no such allowance is made for numbers within the sibships.

To compare the results of using each method, the standard errors in all six color measurements of all nine stocks were computed by both methods and the fifty-four paired values compared as to difference and correlation between the values of each pair. An average difference of .053 color units was found, and since this difference from zero is highly significant it indicates that the standard errors computed by the first method tend to be slightly smaller, on the average, than those computed by the second method. The difference is very small, and the second method tends to minimize differences between stocks. A correlation coefficient of $r = 0.97$ for the fifty-four paired values indicates a high degree of correlation between the estimates of the standard error as computed by the two methods. The second method, basing computations directly upon sibship means, has been used in this paper in making comparisons between stock means (Tables II and III).

Some stocks have the heredity of certain parent mice represented in more than one sibship of the laboratory population. Since the sample of the wild population consists only of the parent mice, some adjustment must be made to avoid falsely augmenting the sample number (n), the number of sibships.

Probably the most desirable method of adjustment is by a system of weights, as suggested by Fisher (1940) or Cotterman (1947) for use in a somewhat similar situation in problems of human heredity. In the present study the adjustment has been made by using as the number of sibships (n) in computing the standard error, a number equal to half the number of wild mice whose heredity contributed to the laboratory stock. In Tables II and III this number has been termed the "effective number" of sibships.

GEOGRAPHIC VARIATION AMONG THE STOCKS

In comparing stock means for significance of differences, the graphical method of Dice and Leraas (1936) has been used. The actual difference between two stocks, with the standard error of the difference, has been computed in only those instances in which the nature of the difference was not shown beyond a doubt by the graphical method. Figures 2 and 3 show the relationship of the stocks in the foot-length measurements and in the dorsal-red readings. These figures will serve as examples of the graphical method as used in both the body and skeletal measurements and the color measurements. The stock means and standard errors used are based upon the distributions of sibship means, as previously explained.

Body and Skeletal Measurements

Variations in mean length of body, tail, and ear among the stocks follow much the same pattern (Table II). In each of the three measurements the variability within stocks is great, and few of the differences between means are statistically significant. The six inland stocks show what may be a tendency for the animals farther inland to be larger than those nearer but not on the coast. Any such cline, if it has any reality, is obscured by the variability of the stock means. The three beach stocks, those from Santa Rosa Island, Destin, and Panama City, seem to be slightly larger than the stocks immediately inland in body length and tail length, but to be about the same in ear length. Again, most of the apparent differences are not statistically significant.

In foot length there is a clear separation between the coastal and inland stocks (Fig. 2). The mean foot length in each of the three coastal stocks is greater than that of any of the six inland stocks. The difference between the stocks from Panama City and Marianna is significant; in all other comparisons between a coastal stock and an inland stock the differences are highly significant. Of the coastal stocks, the Santa Rosa Island mice have the longest feet, the difference from the Panama City animals being highly significant, but neither one of these two stocks differs significantly from

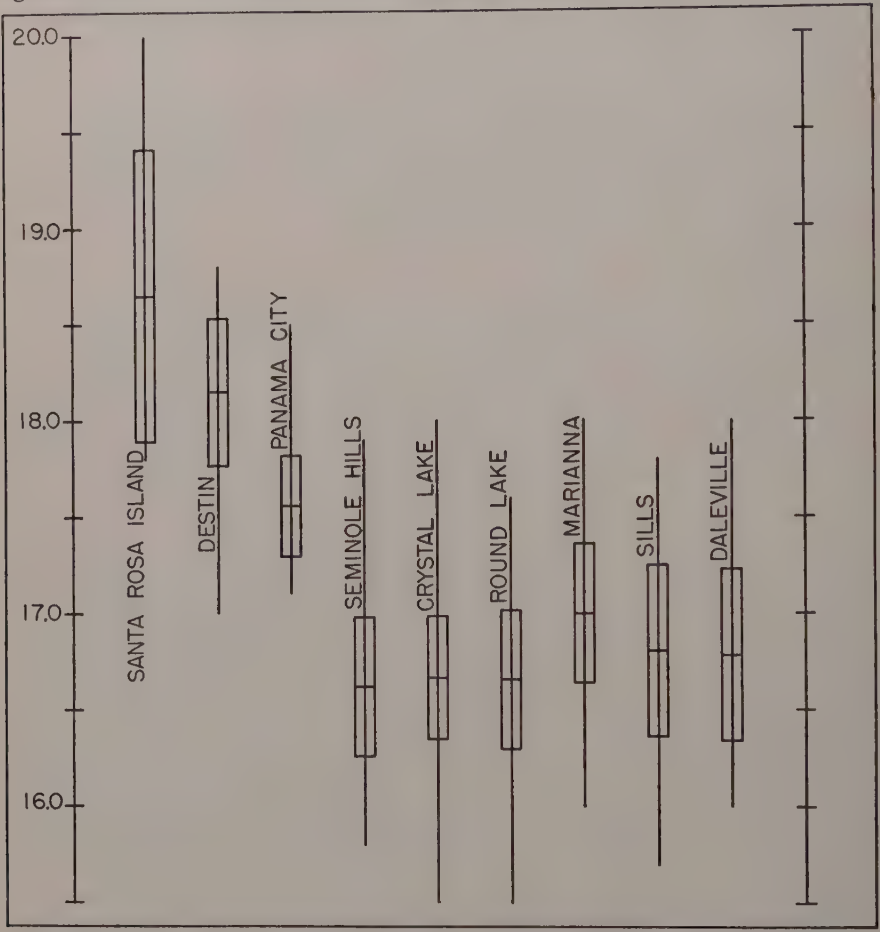

FIG. 2. Variation of hind-foot length of *Peromyscus polionotus*, as shown by the graphical method of Dice and Leraas (1936). Rectangles show twice the standard error each side of the means, which are represented by the crossbars. Observed ranges of measurements are shown by vertical lines. In general, where rectangles do not overlap, the means differ significantly.

the geographically intermediate Destin mice. Among the inland stocks there are no significant differences in foot length. There seems to be a tendency for stocks farther inland to have larger feet than those nearer the coast but not on the beaches, perhaps a reflection of the larger general body size.

Feet which are longer, and perhaps larger in other dimensions, may have some adaptive significance for life on loose beach sands. The Santa Rosa Island mice, with the longest feet, among the coastal stocks, are most completely isolated geographically and also are exposed to the most extreme dune conditions. The Panama City mice, with the shortest feet, are nearest to the inland mice geographically and are presumably the least isolated from them genetically. The Destin mice, with feet of intermediate length, are intermediate in geographical situation and probably also in degree of isolation, as they are at the end of a long peninsula.

In the skeletal measurements used, there seems to be a rough correspondence in the relationships of the stocks from one measurement to another. In femur length, mandible length, condyle-premaxilla length, and condyle-zygoma length, the coastal forms, from Santa Rosa Island and Destin, are the largest, differing in a highly significant manner from the stocks next inland, but being almost equaled by the farthest inland stock, that from Daleville. The Panama City stock agrees in most instances with the other coastal forms, but since it is reduced to a single sibship in the skeletal measurements, it is no longer useful where an estimate of variation is required. In bullar width the same general relationship between stocks seems to hold, though statistically significant differences are fewer.

The six inland stocks, considered apart from those from the coast, may show a general trend toward larger skeletal measurements the farther the animals are from the coast. Any such cline may be reversed in comparing any two collecting stations, but seems to hold in a general way.

In summary, the most noticeable characteristic of the body and skeletal measurements, as Summer (1926) found, is the difference between the coastal and the inland mice. This difference is most apparent in foot length, but is also present in the other measurements, especially in femur, mandible, condyle-premaxilla, and condyle-zygoma lengths, in bullar width, and to a lesser degree in body and tail length. In all measurements there may be a tendency among the six inland stocks toward larger animals farther inland. Such a cline is most apparent in the skeletal measurements and is less clear among the body-measurement data.

Pelage-Color Measurements

The lightest (highest reading) in pelage color of all the stocks is that from Santa Rosa Island (Table III). The readings in this stock are higher than those in any other stock for each of the three colors, both for dorsal stripe and for side. The Santa Rosa Island stock overlaps several of the mainland stocks in the range of the readings, but the differences of the means are highly significant in all comparisons.

The darkest of all the stocks is that from Daleville. The other stocks are scattered in between the Santa Rosa Island and the Daleville stocks, which are not only the lightest and the darkest, respectively, of all the stocks, but are also the farthest apart geographically. Figure 3 illustrates the relationship between the different stocks with regard to the dorsal-red readings. Although the other colors follow the same general trend as dorsal red, there are some differences. The Santa Rosa Island stock will be omitted from the discussion of these differences, since this stock is always much lighter than any other. In the following discussion of pelage color, and in contrast to the procedure in the rest of the paper, only those differences which are significant at the 1 per cent level are noted and termed significant.

In the red readings for the dorsal stripe (Fig. 3) the beach stock from Destin is the lightest. This stock stands significantly higher in readings than any of the other stocks except those from Panama City and Crystal Lake. The Crystal Lake stock is lighter than any of the remaining stocks except that from Panama City. The stock with the next highest reading, from the beach at Panama City, stands significantly higher than any of the darker stocks except that from Seminole Hills. The next darker stock, from Seminole Hills, does not differ significantly from the Round Lake stock. The Marianna stock is significantly darker than the Round Lake stock, but there are no significant differences among the Marianna, Sills, and Daleville stocks.

In the dorsal-stripe reading for green, the stock from Destin is significantly lighter than any of the other mainland stocks except those from Panama City and Crystal Lake. The Crystal Lake stock does not differ significantly from that from Panama City. The next darker stock, from Seminole Hills, is significantly darker than the Crystal Lake stock and significantly paler than the Marianna stock, but is not significantly different from the Panama City stock or the Round Lake stock. The Marianna stock is significantly different from all paler stocks, but none of the differences among the Marianna, Sills, and Daleville stocks is significant.

In the readings of the dorsal stripe through the blue-violet screen, the Destin stock stands the highest, and its difference from any other stock except that from Panama City, is significant. There are no significant differences among the stocks from Panama City, Seminole Hills, Crystal Lake, and Round Lake. Among the three stocks from Marianna, Sills, and Daleville there are no significant differences. Each of these last three stocks is significantly darker than any of the other stocks, except that Marianna and Daleville do not differ significantly from Round Lake.

In the readings of the side stripe for red, the stocks from Destin and

Crystal Lake are paler than any of the others, except that they do not significantly exceed the stock from Panama City. No significant differences exist among the Panama City, Seminole Hills, Round Lake, and Marianna stocks, although all four of these stocks are significantly lighter than the Daleville stock, and all but the Marianna stock differ significantly from the Sills stock. The stock from Daleville is significantly darker than that from Sills.

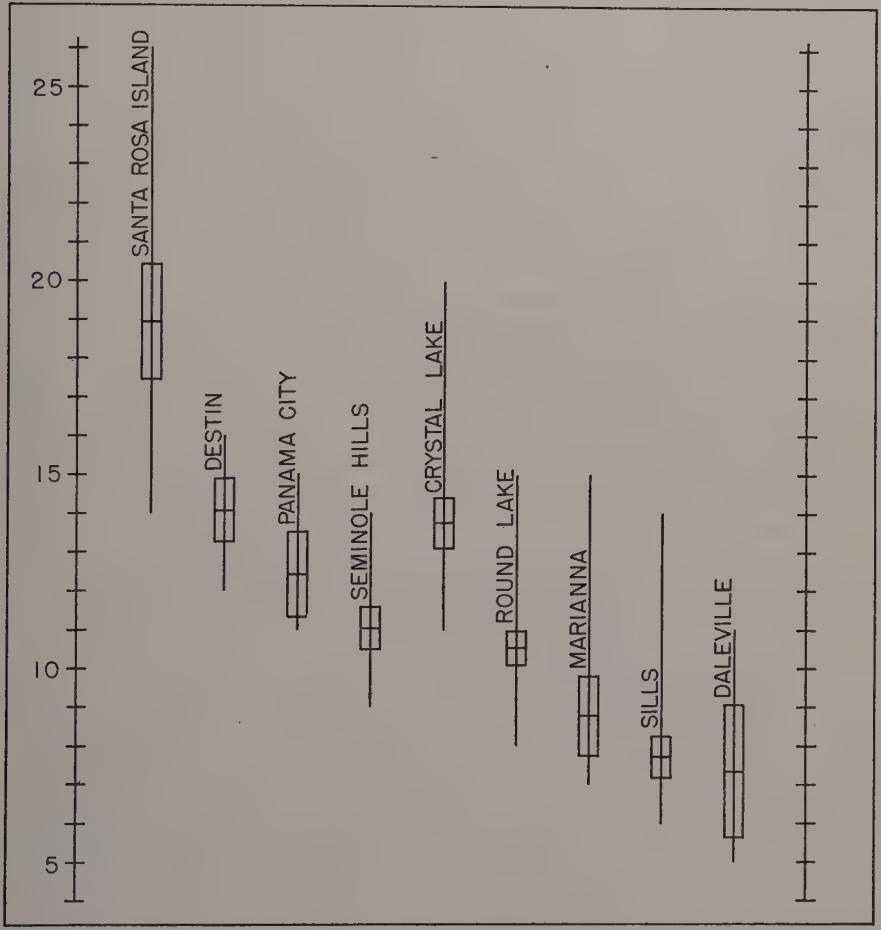

FIG. 3. Variation of tint photometer reading for red on the dorsal stripe of *Peromyscus polionotus*, as shown by the graphical method of Dice and Leraas (1936). See Figure 2 for an explanation of this method.

In the readings for side green, the Daleville stock is significantly darker than all others, and the Sills stock is significantly darker than all except the Marianna stock and the Daleville stock. Among the six remaining inland stocks the only significant differences are between the paler stock from Destin and the darker stocks from Seminole Hills, Round Lake, and Marianna.

With regard to the blue-violet reading for the side stripe, the Daleville stock again has a significantly lower mean reading than has any other stock. Among the remaining seven mainland stocks, significant differences are present in only three comparisons, those between Destin and Seminole Hills, Destin and Marianna, and Panama City and Sills.

Considering all of the color readings together, for the mainland stocks the palest pelage is that of the coastal stock from Destin. It is closely followed by the slightly darker Crystal Lake stock, which differs significantly in only one of the six colors read. The other coastal stock, that from Panama City, stands next in rank of paleness. The Seminole Hills stock is significantly exceeded by the Panama City stock in one of the six color readings. The Seminole Hills stock does not exceed the Round Lake stock significantly in any color reading. The Marianna stock is significantly darker than the Round Lake stock in one of the three dorsal colors, and there is no significant difference in any of the side colors. The Marianna stock does not significantly exceed the Sills stock in any reading. The differences between the Sills stock and the Daleville stock are not significant in any of the dorsal colors, but in the side colors all three differences are significant.

In summary, the two stocks from the light-colored, coastal-beach sand and the stock from Crystal Lake, which lies about in the center of the inland sandy area, tend to be the palest, while the stock immediately back from the beach and that nearest the line between the lighter soil and the darker red soil, those from Seminole Hills and Round Lake, tend to resemble each other and to be somewhat darker than the first three stocks. The stock from Marianna, which is on the dark soils but near the line between the dark and the light soils, closely resembles the Seminole Hills and Round Lake stocks with regard to the side colors but is somewhat darker than they are in the dorsal colors. The differences between the Marianna stock and the other two stocks on the dark soil, those from Sills and Daleville, are greater in the side colors than in the dorsal colors. Thus the Marianna stock may be said to resemble the other two stocks from the darker soil in the dorsal colors but to resemble the stocks from the lighter soil in the side colors.

The results of pelage-color measurements may be compared with the conclusions of Sumner (1929). Both studies record the same tendency for readings through the red screen to decrease as collections were made farther inland. Sumner, however, observed a sudden change in the red readings to take place within a few miles inland from Round Lake, but no such important change occurred in the readings in the present study. The Marianna stock differs in color readings from the Round Lake animals only in dorsal red; indeed, in the color readings for side stripe the two stocks are almost identical. Inclusions in the present study of the Seminole Hills stock, which is composed of darker animals than either the coastal stocks or that from Crystal Lake, destroys the appearance of a uniform gradient between the beaches and Round Lake.

Detailed comparison of color measurements made in the two studies cannot be made, since the single reading of Sumner is equivalent to an approximate average of the two readings of the present study. Nevertheless, an approximate comparison may be made, using data from the two points where the collecting stations of the two studies coincided. At Crystal Lake Sumner's value for red (17.3) agrees well with the two values of the present study (dorsal red, 13.7; side red, 20.3). At Round Lake Sumner's value for red (15.7) may slightly exceed the corresponding values of the present study (dorsal red, 10.6; side red, 17.0). This agreement indicates that the observed differences in geographic trend probably do not arise from different methods, but are the result of collecting at different localities.

In addition to his use of tint-photometer readings, Sumner compared pelage characteristics by a number of measurements not carried out in the present study. Several of these measurements are closely related to the more obvious differences between the two subspecies *albifrons* and *polionotus,* and without doubt would show a sudden change in the area between Round Lake and Marianna, where the change in described subspecies occurs.

Correlation Between Pelage Color and Soil Color

Characteristics of pelage color have been used to an important extent in dividing the animals of the species *polionotus* in this area into three subspecies (Osgood, 1909; Howell, 1920). There is no difficulty in assigning the animals of any of the nine stocks of the present study to these three races. The extremely pale animals from Santa Rosa Island are of the subspecies *leucocephalus.* Five stocks are assignable to the subspecies *albifrons,* including the two beach collections from Destin and Panama City, and the three inland stocks from Seminole Hills, Crystal Lake, and Round Lake, these last three being named in order of closeness to the beach. The three remaining stocks have the characteristics of the subspecies *polionotus* and, again in order of closeness to the beach, are those from Marianna, Sills, and Daleville (see Fig. 1).

When the mice are compared according to subspecies, there is a strong correlation between pelage color and soil color (Table XVI). Mice of the subspecies *leucocephalus,* by far the palest in pelage, are present on the extremely pale sands of Santa Rosa Island. Mice of the subspecies *polionotus* are darkest in pelage and are found upon the dark soils of the interior. Mice of the subspecies *albifrons* are intermediate in color readings and are present on soils which are intermediate in average color readings. Table

TABLE XVII

PELAGE COLOR AND SOIL SAMPLE COLOR FOR RACES OF *Peromyscus polionotus*

The first figure is the mean pelage-color reading; the second figure, within parentheses, is the mean soil-sample reading.

Race	Number of Stocks	Dorsal Colors			Side Colors		
		Red	Green	Blue-violet	Red	Green	Blue-violet
leucocephalus	1	19 (65)	15 (61)	14 (58)	27 (65)	22 (61)	19 (58)
albifrons							
All stocks	5	12 (43)	9 (38)	7 (35)	18 (43)	14 (38)	11 (35)
Coastal	2	13 (64)	10 (61)	8 (58)	19 (64)	15 (61)	12 (58)
Inland	3	12 (29)	9 (23)	7 (19)	18 (29)	13 (23)	11 (19)
polionotus	3	8 (24)	6 (16)	5 (10)	14 (24)	11 (16)	9 (10)

XVII lists the average color readings for soil and pelage, when the stocks are grouped according to subspecies. These data are useful for only the most approximate of comparisons, since neither the soil samples nor the mouse stocks are random samples of the color relationships throughout either the subspecies or the geographic area.

When comparisons are made within the subspecies, then correlation of pelage color with soil color becomes much less apparent. Among the *albifrons* stocks the mice from the two locations on the pale sands of the Gulf beaches are paler than the average of the three *albifrons* stocks from inland points, and hence produce the appearance of a slight correlation. Among the *polionotus* stocks variation in pelage color and soil color seem to be almost at random within the limits of the subspecies.

TABLE XVIII

TOTAL AND PARTIAL CORRELATION COEFFICIENTS IN *Peromyscus polionotus*

Variable y is pelage color, variable 1 is distance from the coast in miles, and variable 2 is soil color.

Color Reading	Total Correlation Coefficients			Partial Correlation Coefficients	
	Pelage Color and Distance r_{y1}	Pelage Color and Soil Color r_{y2}	Soil Color and Distance r_{12}	Pelage Color and Distance, Allowing for Soil Color $r_{y1.2}$	Pelage Color and Soil Color, Allowing for Distance $r_{y2.1}$
Dorsal Red	–.89**	.65	–.70	–.80*	–.09
Dorsal Green	–.88**	.67	–.72*	–.78*	.11
Dorsal Blue-violet	–.92**	.83**	–.78*	–.77*	.46
Side Red	–.64	.50	–.70	–.47	.10
Side Green	–.85**	.52	–.72*	–.81*	.26
Side Blue-violet	–.83**	.62	–.78*	–.71	–.09

* Difference from zero significant.

** Difference from zero highly significant.

The eight stocks of *albifrons* and *polionotus* grouped together without regard to subspecies may be compared as to pelage color. Each stock mean is here used as a single value. The eight values have been used in computing the correlation coefficients for pelage color, soil color, and distance of the collecting locality from the coast (Table XVIII). Distance from the coast is not used here as necessarily of any direct biological significance, but merely as an approximate measurement of whatever factors are effective in producing the obvious gradation in pelage color of specimens, as collections were made farther inland (Fig. 4).

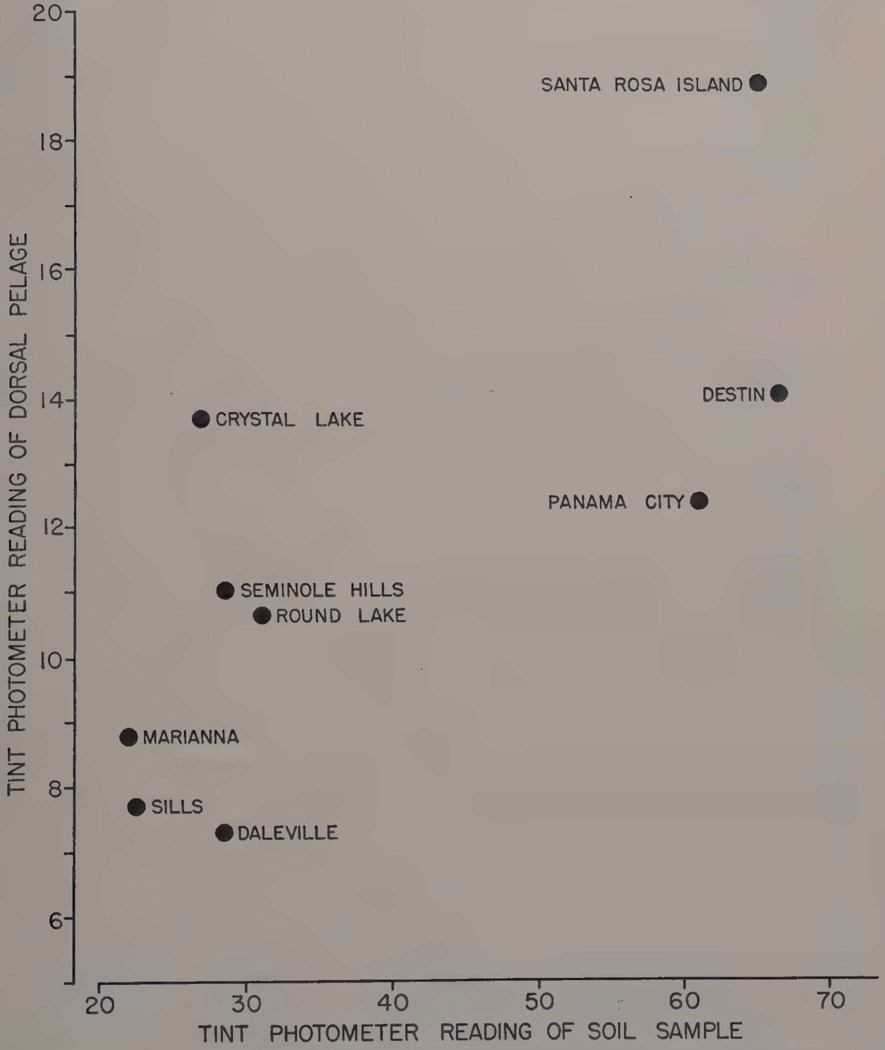

FIG. 4. Relationship between dorsal pelage readings of *Peromyscus polionotus* and soil sample readings, for red.

Use of a correlation coefficient implies a linear relationship between the variables involved. From the present data it seems reasonable to assume a linear relationship between soil color and pelage color (Fig. 4). But the relationship between pelage color and distance from the coast does not seem to be necessarily linear; judging from Sumner's results, it should not be expected to be.

The data of this study, however, even though variable, do seem to conform closely enough to a linear relationship (Fig. 5), so that a correlation technique may be used in an approximate description of the observed cline.

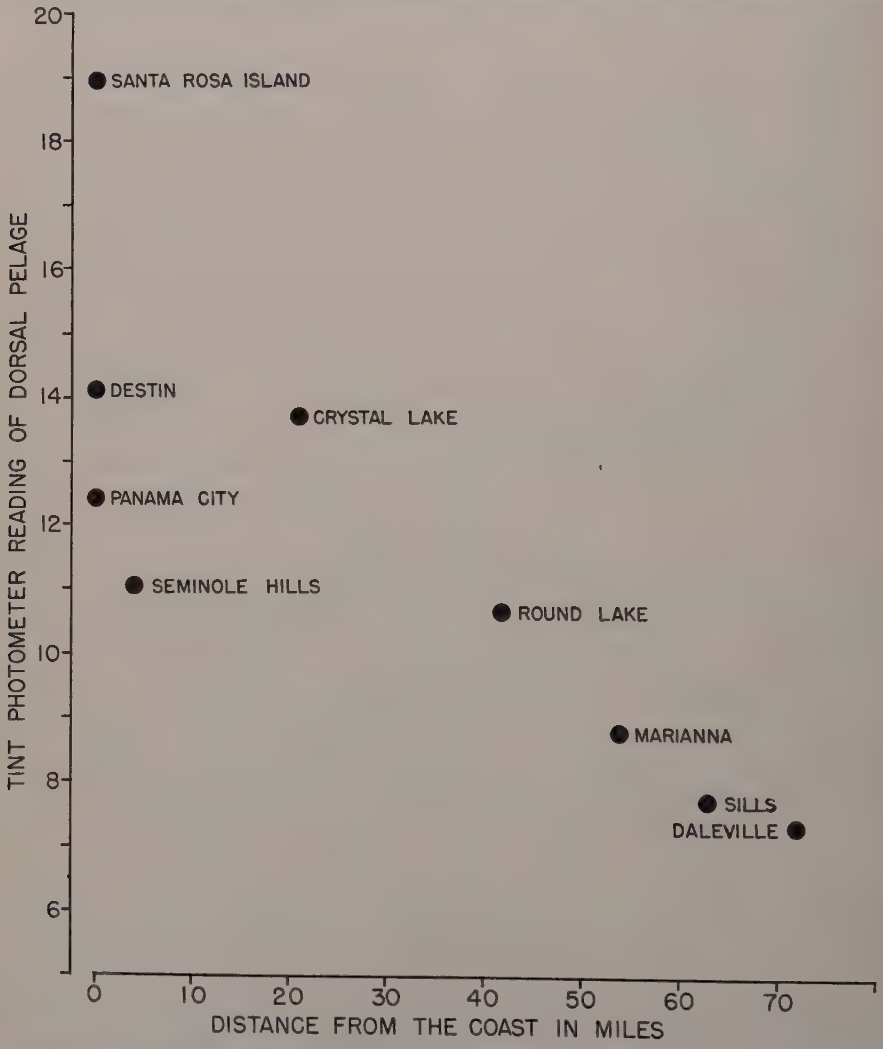

Fig. 5. Relationship between dorsal pelage readings for red, and distance from the coast in *Peromyscus polionotus.*

The term "total correlation" is used to define the apparent correlation between two variables without reference to any other factors. The term "partial correlation" is applied to the correlation of one variable with a second, when allowing for the effect of a third variable which may be correlated with both of the other variables (Simpson and Roe, 1939; Snedecor, 1946).

The values of the coefficients of total correlation (Table XVIII) indicate a strong relationship in most instances between pelage color and distance from the coast, but a less clear correlation between pelage color and soil color. When allowance is made for distance from the coast (by means of partial correlation), the correlation of pelage color and soil color becomes negligible. Correlation of pelage color with distance from the coast, however, is only slightly reduced by allowing for soil color. Mathematically, soil color would be far less reliable than distance from the coast in predicting pelage color under the particular conditions of this comparison.

The apparent importance of distance from the coast seems to be the biological result of the action of two factors, (1) the matching of the pelage hue to the soil background and (2) the genetic interchange due to the dispersal of breeding individuals from any population to adjacent areas. The three chief soil areas with regard to soil color in this area lie roughly parallel to the coast, and the intergradation across the three belts of soil produces a fairly uniform gradient in pelage color, even though the gradient in soil color is nonuniform. The effect upon pelage color of the exact local soil color seems to be obscured by the effect of intergradation.

When these animals are considered according to subspecies, there is a strong correlation between pelage color and soil color. It has been reported in a number of studies that small mammal pelage color tends to match soil color (Dice, 1930, 1939, 1940*a,* 1941, 1942; Benson, 1933; Dice and Blossom, 1937; Blair, 1941). The present correlation appears to bear this out. That the correlation is so strong when the stocks are grouped according to subspecies appears largely to reflect the use of pelage-color characteristics by taxonomists in setting up these subspecies. Within any of these pelage-color races, however, interbreeding, the spread of hereditary material through the race, appears to be important in determining the average color of a local population, so that any local variations in soil color appear to have little effect upon the precise pelage color within the local population of mice.

DISCUSSION

The effect of various factors in altering or maintaining the genetic variability within populations has been considered from the theoretical aspect by Wright (1931, 1943, 1946, and numerous other papers), and his ideas have been discussed by other workers in various fields, including Dobzhansky

(1941) and Huxley (1942) in relation to geographic and ecological variation, and Lush (1945) in relation to animal breeding.

The "factors of evolution" have been listed by Wright (1931) according to their effect in increasing or decreasing genetic variability within a population. Among the more important factors tending toward genetic homogeneity are (1) random fixation or loss of genes within a population, which may be especially effective when the breeding population is restricted in number, and (2) natural selection operating to favor the survival of one gene over its allelomorph. Among the factors tending toward genetic heterogeneity are (1) gene mutation and (2) hybridization between breeding populations, especially as brought about by the carrying of genetic material into a local population from an outside source such as a neighboring population. Each of these factors will be examined as to its possible relation to variability in the stocks of the present study.

The effective number of breeding animals in the local population is independent of the particular characteristic chosen for study, and this number of breeding animals is the same for all characteristics at any one time. The effect of breeding-population number upon genetic variability may be influenced by mutation rate, by selection, or by the amount of new genetic material being brought into the population from outside. Wright (1931) noted that in the case of very small populations, the genetic variability will be eliminated at such a high rate that mutation and selection can have little effect.

Population number is not known for any of the stocks in the present study, but the generally low level of genetic variability in the color data in most stocks does not seem due solely to genetic drift in very small populations. In the same stocks the body-length data often show a significant degree of genetic variability. Variability in three stocks is not highly significant in any of the color readings, in body length, or in foot length. These stocks are from Santa Rosa Island, Panama City, and Daleville, and only in these three localities were the data consistent with a hypothesis that the low genetic variability may be due to genetic drift in very small populations.

Hybridization has an effect opposite to that of restricting the size of the breeding population. A high rate of interchange of genetic material between local populations tends to maintain the genetic variability. Wright has considered a number of special cases of this problem, including isolation, the introduction of genes from the entire species population (1931), and local inbreeding in a continuous population, with "isolation by distance" (1943; 1946). He has used the two terms "migration" and "dispersal" to describe the movements of individual animals between local populations, resulting in genetic interchange. "Dispersal" seems preferable, since "migration," as used by Wright, carries a somewhat different meaning than the same term does when used by certain animal ecologists. Heape (1931) restricted the meaning of migration to those movements in which an animal is impelled to return to the region or to the local area from which it came. In dispersal, as the term is used in the present study, the movement is away from a local population or homesite and random in direction as to other local populations, though not necessarily random as to habitat or ecological community. Whether such a movement is involuntary, as Heape would define it, or whether the movement may be voluntary, has little relation to the present problem. Wright's designation of an animal as an ''immigrant'' after it has entered a local breeding population from an outside source seems appropriate. An immigrant according to this usage, would be an animal which had participated in a dispersal from its original home area.

The number of immigrant animals entering a local population within any interval of time will be an aspect of the local population and its ecology. Dispersal rate, like population number, will have a local numerical value, whatever the measurement being studied. Any genetical effect of dispersal is a result of the immigrant genetic material being different from that of the local population. When there are no genetic differences between local populations, genetic interchange as a result of dispersal can only have the effect of increasing the size of the breeding population. Where hereditary differences are present, genetic interchange will raise the genetic variability. The effect of dispersal will, therefore, depend upon the degree of genetic difference betwen populations which exchange breeding individuals, and this difference in hereditary material may vary from measurement to measurement.

The rate of dispersal will have some relationship to physical and ecological barriers. Rate of dispersal and its relative importance are unknown for the populations in the present study, but there appear to be certain barriers to free movement. The waters about Santa Rosa Island constitute the only apparent physical barrier to all movement of mice. Although this isolation prevents genetic interchange between island and mainland animals, it is not necessarily the sole cause of the low genetic variability of the Santa Rosa Island animals. The fifty-mile length of Santa Rosa Island seems to give ample opportunity for the development of local populations.

Barriers which at least appear to reduce free dispersal are present at other places. The narrow zone of marsh behind the dunes at the Panama City location would seem to decrease genetic interchange between the populations of mice on the beaches and those in the flatwoods a mile or two inland. Any such check on dispersal would aid in maintaining the differences between the coastal stocks and those immediately inland in pelage hue and in foot length. The location of Destin on the tip of a peninsula probably limits the number of local populations with which an exchange of breeding animals can be carried on. The population at Sills is completely surrounded by an expanse of wooded swamps; yet the presence of the animals in this isolated field probably indicates that they came there originally through the apparent barrier. In this stock there is evidence of genetic variability in body length, hind-foot length, and in the readings for red on the side stripe.

The hypothesis that some genetic exchange occurs between local populations is supported by the evidence of genetic variability in factors controlling pelage hue in the two stocks from Round Lake and Marianna. Genetic interchange in this area, in which the subspecies *albifrons* and *polionotus* meet, should be expected to maintain a high rate of variability, because the two subspecies differ markedly in pelage hue. Natural selection, discussed farther on, may also increase variability in pelage hue in this area. In the foot-length data, which do not differ markedly between the two subspecies, the Round Lake and Marianna stocks have no higher variability than the other stocks.

Mutation rate as it affects the various stocks is an unknown factor. It is not necessary to assume either a constant mutation rate or any particular differential rates among the stocks or characteristics studied. Mutation is undoubtedly important, but nothing is known of its effect in this species.

Natural selection results in a tendency for one combination of hereditary traits to survive at a higher rate than another combination. Wright (1932) illustrated the effect of natural selection with a model consisting of peaks and valleys, in which a peak represented the optimum value of a characteristic. A population subjected to severe selection will be held near the optimum value, near the top of a peak, and much of the genetic variability will be eliminated. In a population in which selection is at a low rate, the genetic variability will be less severely eliminated, and the value of the population mean may "wander" about the optimum value, or in terms of the model, the population mean may wander down on the slope surrounding the peak.

Natural selection was apparently important in the evolution of local pelage-color races in the area of the present study. The major changes in soil color were accompanied by corresponding shifts in pelage hue. The low genetic variability in all stocks, except the two from Round Lake and Marianna, is in agreement with an hypothesis that selection is operating upon pelage-color characteristics.

The relatively high degree of genetic variability in pelage hue in the stocks from Round Lake and Marianna may arise from two sources. First, the stocks are in the area of meeting of the two subspecies, which are quite dissimilar in pelage color, and any given degree of genetic interchange between local populations will result in a greater increase in variability than would occur in an area wholly within the range of either subspecies.

Second, the cause of greater genetic variability in the two stocks in this area may be natural selection. In the region between Round Lake and Marianna, especially immediately north of Round Lake, the soil color is variable, interspersed with patches of dark- and light-colored soils. Wherever selection favors the animal which most nearly matches its immediate background, two neighboring local populations might be selected in opposite directions so far as pelage hue is concerned. Even within the same breeding population, selection might eliminate darker individuals exposed on pale soil and at the same time eliminate paler individuals exposed upon dark soil. This situation resembles the theoretical one discussed by Fisher (1930) and Wright (1931), in which selection favored the survival of the heterozygote in preference to either homozygote (in the case of a single pair of genes). Under such conditions natural selection would not decrease the genetic variability.

In body length, selection appears to have been less important in the development of local races. As shown by the differences between sibships, none of the beach stocks and all of the inland stocks carry a significant degree of genetic variability in this character. What possible adaptations may be involved in the differences between stocks is not known. Few of the differences are statistically significant as judged by these samples.

In foot length, the three coastal stocks are clearly separated from the inland stocks in that the coastal mice have longer feet and show no genetic variability in foot length within stocks. If a longer foot is an adaptation to life on beach sands, then selection may account for the low variability within the beach stocks. In the inland stocks a significant or highly significant degree of genetic variability is present in five of the six stocks.

In pelage color, the gradient in the area of the present study seems to be the result of the intergradation of two, or possibly three, color races, each matching roughly the color of the soil in its own geographic area. In the area in which *polionotus* and *albifrons* meet, each race shows the effect of the presence of the other in a gradient of pelage color extending on each side of the line of meeting. Possibly near the beaches the pale beach populations show a similar intergradation with the somewhat darker adjacent inland populations, which are classified as the same subspecies. The two gradients in pelage color are in the same direction, so that the appearance of a more or less uniform cline over the whole area is produced.

The pelage hue of any one population apparently represents a balance between the tendency for natural selection to decrease the genetic variability, to match each population more precisely to its immediate background, and to narrow the area of intergradation between markedly different races, and the opposing tendency for the dispersal of breeding animals to maintain the genetic variability, to make each population more like an average of all populations in the area, and to broaden the area of intergradation. The precise mean pelage color in a population may also ‘‘wander’’ due to the sampling nature of breeding, and the investigator introduces a final source of variation in his sampling error.

The paper by Haldane (1948) treating the theory of a cline has come to my attention too late to be adequately used in the present study. With a mathematical treatment Haldane has related the opposed effects of selection and dispersal and shown how a cline may result from a balance between these two factors. Using Sumner's data for change in "colored area" in mice of the region of the present study, and making certain assumptions, Haldane has shown that even the pronounced gradient observed in this characteristic could be maintained by natural selection, operating to produce an advantage of about one-tenth of 1 per cent, on each side of a boundary.

SUMMARY

Nine populations of the oldfield mouse, *Peromyscus polionotus,* in northwestern Florida and southern Alabama have been sampled by capturing breeding animals in the field and measuring certain characteristics in their laboratory-bred offspring. These stocks are representative of three described subspecies, *leucocephalus, albifrons,* and *polionotus.*

In the type of sampling here employed, the true sample of the wild population is the wild-captured animals. Because of the necessity of subsampling by breeding the wild-captured animals, the laboratory-bred sibship becomes the actual unit of the sample. The mean of the wild population is best estimated by the mean of the sibship means. A standard error must be based upon the number and variability of the sibships which compose the sample. The standard deviation of the wild population is estimated moderately well by the standard deviation of all laboratory offspring considered without regard to sibship.

The most efficient division of the laboratory population into sibships and offspring within sibships can be computed for any particular ratio of hereditary to nonhereditary variability. Because of the importance of numbers of sibships in sampling the inherited variability in a population, the safest plan seems to be to sample the wild population with as large a number of sibships as possible.

Significant differences between sibships within a stock are interpreted as indicating a significant degree of hereditary variability within that stock for the particular measurement. The same stock may show significant differences between sibships for one characteristic, with no differences between sibships for another characteristic, an indication that the degree of genetic heterogeneity may vary from characteristic to characteristic.

Body-length data have a highly significant or a significant amount of genetic variability in five of the six inland stocks and in one of the three beach stocks. The body and skeletal measurements may possibly show a tendency among the animals of the inland stocks to be larger the farther

they are from the coast. Three coastal stocks seem to be made up of larger animals than those stocks immediately inland. Any geographic cline in body or skeletal measurements which may be present is obscured by the considerable variability of the samples.

The beach mice have longer feet than do animals from the inland areas. The longer feet may have some adaptive value upon beach sands. No significant genetic variability is noted in any of the beach stocks, but of the six inland stocks, five show highly significant or significant variability. These observations are consistent with natural selection with regard to foot length being more important upon the beaches than it is inland.

Pelage-color measurements indicate that genetic heterogeneity is relatively unimportant in all the stocks except the two from Round Lake and Marianna. In these two, the important proportion of the variability which is inherited may be the result both of dispersal in a locality where two races meet and of natural selection acting against both pale and dark pelage hue in an area interspersed with small areas of contrasting soil colors.

The color measurements show an apparent cline in pelage hue, and this appears to be the result of intergradation among (*a*) the darker mice from the red soils farthest inland, (*b*) the paler mice from the buff and gray soils between the red lands and the beaches, and (*c*) the still paler animals of the Gulf beaches.

Over the whole area of the study there is a strong correlation between pelage hue and soil hue. From place to place within any of the three important soil-color areas, however, there appears to be little or no correlation between the average pelage hue and the local soil hue.

Natural selection and dispersal appear to be important factors in the determination of both the mean values of the measurements and their variability within local populations. Genetic drift due solely to small population size seems unlikely to have been important in reducing variability in the several populations studied.

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